PHYTOPLANKTON OF SHEARWATER WITH SPECIAL CONSIDERATION OF FUNGAL PARASITES AND EPIPHYTES

A thesis submitted for the Degree of Doctor of Philosophy at the University of Bristol

by

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MEMORANDUM

The work reported in this thesis is the result of my own independent research under the supervision of Professor F.E. Round, University of Bristol.

Bülent Sen.

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SYNOPSIS

The phytoplankton of Shearwater, Wiltshire has been studied over three years paying particular attention to the epiphytic populations on the individual algal species. In order to relate any effects of the epiphytes on the seasonal growth of the algae it was necessary to monitor the physico-chemical factors and compare the cycles of individual algae and groups of algae with these factors. Unlike many more oligotrophic lakes, Shearwater maintained high populations of phytoplanktonic algae throughout the whole study period. Individual species also maintained their growth over longer periods than expected. Correlations between phytoplankton and physico-chemical factors were made wherever possible.

Epiphytic populations of fungi (mainly Chytrids) protozoa and other algae were continually monitored on the phytoplankton. In the case of the Chytrids, this also involved studies of stages in their life cycles in order to identify the fungi. Their effects on the growth patterns of the algae were documented.

There is no literature on the occurrence of choanoflagellates on algae and hence the present study of these in Shearwater provides unique data.

Data on the epiphytic populations was obtained by a combination of light and electron microscopy.

The taxonomic position of the epiphytes and their relationship to the phytoplankton is discussed with reference to the literature.

One new fungal species is described.

CONTENTS

INTRODUCTION

A considerable amount of information has been gathered since the beginning of the present century in limnological studies of many waters of the world. The bulk of these studies have invariably concentrated on the seasonal periodicity of phytoplankton, primary productivity and water chemistry. Physico-chemical factors have generally been considered primarily responsible for the seasonal cycle of planktonic algae, consideration of biological factors, such as parasitism and interaction between organisms being largely ignored. In fact, very few investigations have been made hitherto on the effect of parasitic fungi on phytoplankton populations despite the fact that pioneer works of CANTER & LUND (1948, 1951) showed that these organisms could exert an important influence on the cycle. Algae form the major part of phytoplankton and include both eucaryotic and procaryotic organisms which may be parasitized by viruses, bacteria, fungi and protozoans. In fact, organisms in aquatic environments and indeed in other communities are interdependent in many ways and their distribution should not be interpreted without considering their mutual inhibitory and stimulatory effects. The present work comprises a comprehensive study of the effects of fungal parasitism together with physico-chemical factors on the seasonal cycle of phytoplankton population in a freshwater lake. It is also well known that some algae, bacteria and protozoans often occur on algal cells or in the mucilage surrounding them. It is yet not known to what extent these non-parasitic micro-

organisms affect the growth of algae. Shearwater - a freshwater lake investigated in this study - has provided good examples of this kind of micro-association during this study. The ecology of epiplanktonic organisms such as Stylosphaeridium stipitatum and members of the zoomastigophoran genera Bicosoeca, Codosiga and Salpingoeca has been studied quantitatively for the first time and the role of planktonic algae in harbouring significant populations of these organisms has been considered. In order to assess these relationships, it was essential to fully understand the seasonal periodicity of the representatives of the phytoplankton.

CHAPTER I.

INTRODUCTION TO SHEARWATER - METHODS & PHYSICO-CHEMICAL DATA

Shearwater is a small, artificial lake in Wiltshire (Map ref. ST 850421) lying on Cretaceous Upper Greensand. It lies in a catchment of deciduous woodland and agricultural land and is therefore richly supplied with nutrients from small streams. It is used for recreation, mainly sailing. No detailed survey of the lake has been undertaken and only three papers on its algal flora have been published. ROUND (1965) studied the epipsammic flora of the sandy sediments, MOSS & ROUND (1967) reported on the standing crops of the epipelon and epipsammon and HICKMAN & ROUND (1970) studied the primary production of the epipelon and epipsammon over a two year period. The rich phytoplankton has never been sampled before and so this is a first account both of the phytoplankton, its associated epiphytes and parasites and the chemical status of the water. Sampling was from a wooden platform built out into the water for use of the sailing club and hence all the data has been obtained on sub-surface populations close to the shore. No information is available on changes with depth or of the sequence of stratification of the water. The flora clearly indicates a eutrophic lake with a distinct seasonal cycle of spring diatoms, summer Chlorophyta and autumn Cyanophyta. The continual drainage and outflow over a weir maintains a relatively high nutrient supply throughout the year and the phytoplankton populations also tended to be high throughout the sampling period.

ECOLOGICAL STUDIES

The literature on the ecology of freshwater phytoplankton has now become columinous with the continuous studies of algal periodicity and the factors determining the ecology of phytoplankton. Many factors may influence the development of an algal bloom , the duration of a bloom and the ultimate decline of a bloom in freshwaters. It is, therefore, difficult to review the effects of each environmental factor separately. The subject is so diverse that it cannot be dealt with on a simple fact to fact basis, but requires information from many fields of interest in order to understand this complex system. Some physical factors are very complex but it is usually easier to investigate the effects of physical factors than those of chemical factors. Biological and chemical factors merge into one another in the field of organic chemistry (e.g. extra-cellular and inhibitory substances). Nevertheless, field studies and data on the chemical composition of the aquatic environment are indispensible for the ecological studies.

METHODS AND FIELDWORK

Shearwater was visited as regularly as possible every fortnight from April 1977 to February 1981 but occasionally it was not possible to do the regular samplings due to adverse weather conditions and inavailability of transport. However, monthly visits were made instead during such periods.

During adverse conditions of the 1978/1979 winter, the sampling was possible after breaking the thick ice which covered the surface of the lake. Phytoplankton sampling and water samples on which chemical analyses were carried out were collected at the same time.

Algal Sampling and Estimation

Surface phytoplankton samples were taken by means of a fine nylon net which was thrown out as far as possible and then drawn in. During high density of phytoplankton, the samples were collected by repeating this procedure 2 - 3 times. The samples were then placed in screw-top jars which were previously washed with the lake water. Precaution was taken not to collect very concentrated samples and fill up the jars in order to reduce the decomposition risk.

On the return to the laboratory, phytoplankton sample was examined immediately and it was then used either for electron microscopy or kept in the refrigerator for further examinations. A volume of sample was also preserved with a saturated solution of iodine in potassium iodine or with 4% gluteraldehyde solution for later use.

A second sample for algal counts was taken by slowly immersing a bottle below the surface of the water. The algal numbers were obtained by sedimentation of 1 ml of suspension of the sample with 1 drop of saturated solution of iodine in potassium iodine (Lugol's solution) counting on an inverted microscope by the LUND et al (1958) technique. The cells and

colonies were allowed to settle down for 3 - 4 days in a cylindrical glass tube, 2.5 cm high and 1 cm in diameter. The whole bottom of the tube was examined in order to obtain an accurate count of individual algal cells or colonies and this was repeated three times. The mean number of those replicas was then expressed as number of cells or colonies per ml. of water. Any samples with a high density of algae were diluted until a suitable distribution of algae for accurate counting was obtained.

With regard to coenobiate planktonic algae such as Pediastrum, Scenedesmus, the number of cells was recorded. Each coenobium was recorded as a single unit for Pandorina, Dictyosphaerium, Coelastrum, since an accurate count of cells was not always possible. It was also difficult to count the cells of Fragilaria, Melosira and the planktonic blue-green algae such as Anabaena, Aphanizomenon, Coelosphaerium, Gomphosphaeria and Microcystis, thus each colony or filament were considered as a single unit.

Physico-chemical analyses and Estimations

Waters for chemical analyses and physical measurements were collected drom the point at which surface temperature was carried out. Samples were brought back to the laboratory in polythene containers and analyses carried out as soon as possible to minimise any possible chemical changes. The water for chemical analyses was filtered immediately through Whatman filter paper in order to eliminate all micro-organisms including

bacteria which otherwise interfere with the accuracy of the determination.

Hydrogen-ion (pH) concentration

This was measured by using Electronic Instrument Ltd. pH meter 7020. Before any measurements commenced, the machine was standarized using a buffer solution of pH-7 and the temperature of water sample and buffer solution was adjusted to that of the lake at time of sampling.

Surface temperature

Surface temperature was recorded at the times of sampling using an ordinary Centrigrade thermometer. Measurements were carried out in the mornings, usually between 10.30 a.m. - 11.30 a.m.

Water level

Water level was recorded by means of a metric ruler from a permanent point.

Nitrate

The phenol disulphonic acid method was employed for nitrate estimations (Latest FBA method). After evaporation of the sample to dryness, the nitrate present reacts with

concentrated phenol disulphonic acid to produce a yellow nitrated derivative. Interference from colouring matter was reduced by absorption onto magnesium hydroxide and absorbance determined spectrometrically after dilution. Estimations were made as mg NO_3 - N/l.

Phosphate

The method, modified from MURPHY & RILEY (1962) and STEPHENS (1963) was used for phosphate determinations. Phosphate reacts with molybdate to form molybdo-phosphoric acid, which is then reduced to the intensely coloured molybdenum blue complex and determined spectrophotometrically. Increased sensitivity is obtained by extracting the blue complex into an organic solvent (hexanol). Estimations were expressed as mg PO_4-P/l.

Silicate

Dissolved silica determinations were done by the reduction of silico-molybdate complex by metol-sulphite solution acidified with oxalic and sulphuric acids (MULLIN & RILEY, 1955). Absorbance was determined spectrometrically and the results were expressed as mg Si/l.

PHYSICO-CHEMICAL FEATURES OF SHEARWATER

Physico-chemical measurements are necessary in order to provide the necessary basic information to relate to the biological activities in a lake. In addition, the trophic status varies from season to season hence continual measurements were made alongside the seasonal fluctuations of the species. Thus the occurrence of a species could be correlated with physico-chemical data.

pH:

The hydrogen-ion concentration in the aquatic environment may act on phytoplankton as a controlling or a lethal factor, according to its level. The pH may control the rate of some enzyme activity within the algal cell and the lethal effect will occur when the pH reaches a value outside of the pH-tolerance limits for the alga. Some algae appear to be adopted to alkaline conditions while other species occur under acidic conditions.

pH values measured in this study ranged between 7.3 - 7.8, showing that Shearwater is a slightly alkaline lake (Fig. 1.). The general range was usually 7.4 - 7.7 and slightly lower and higher values were recorded on some occasions. The highest values were achieved in April and June - July 1978, while in 1979 May and August - September comprised the highest pH values. The lowest values occurred in October 1978 and February and November in 1979. Changes in

pH values were inconsiderable and pH did not show a clear seasonal pattern from year to year in Shearwater. Nevertheless pH tends to increase sometime in Autumn and reaches its highest value in Spring.

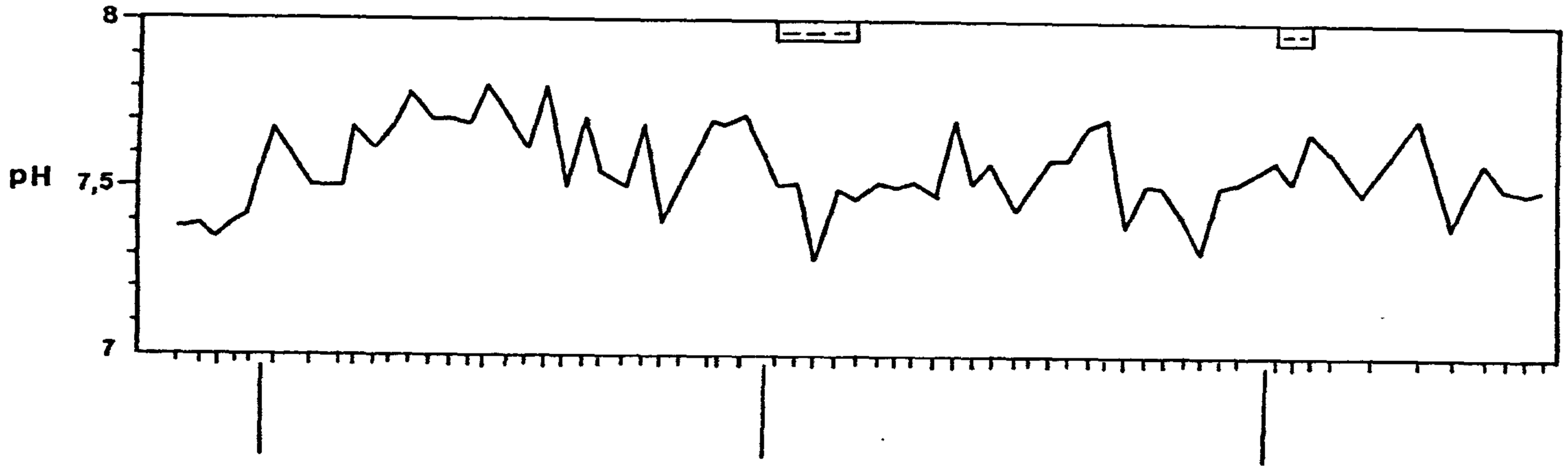
The highest pH values coincided with low water levels while the lowest measurements were synchronous with high water levels in Shearwater (Figs 1 & 2). However it is noteworthy that rise or fall in pH did not follow those of water levels.

With regard to its relation with temperature, the highest pH synchronized with increasing and high temperatures while the lowest values were recorded when temperature was decreasing in Shearwater (Fig. 1.). Ice formation appeared to affect the pH since there were either sharp or slight decreases in pH when Shearwater was covered by ice in two consecutive years. However pH increased or declined regardless of the temperature in this study. The rise in temperature reduces the solubility of carbon dioxide and this may in part account for the increased pH. It is also well known that a heavy growth of phytoplankton can raise the pH of the environment when the increased photosynthetic demand reduces the carbon dioxide content of the water. For example, CHAMBERS (1915) noted that phytoplankton blooms raise the alkalinity of the water and PEARSALL (1930), WADE (1949/50) and RAO (1955) related the rise in pH to increased photosynthesis.

The influence of pH on the growth of planktonic algae is particularly complicated since the pH of an environment depends upon the levels of other factors. Nevertheless, seasonal cycles of planktonic algae in Shearwater with regard to changes in pH

Fig.1. Seasonal variation in pH (upper) and temperature (lower graph)

⊏=⊐ indicates the ice period in Shearwater. The same symbol is used for this period in Shearwater throughout this study.


8
pH
7,5
7


20
15
°C
10
5
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F
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1978
1979
1980
1981

will be highlighted in later chapters.

Temperature:

Temperature is of paramount importance in limnological studies since it has an obvious effect on the physiology and growth of organisms and also affects the environment. Different species undoubtedly have different temperature requirements and rise or fall in temperature might affect adversely one organism while favouring the development of another. It is impossible to separate temperature from light because of their inter-relations in photosynthesis. However, only the surface temperatures of Shearwater were measured in this study. The occurrence of members of the phytoplankton can be correlated only with this.

Fig. 1. shows the measurements of surface temperature in Shearwater. It is apparent from the same figure that temperature showed a very similar pattern with little differences in each year, over the periods studied. Surface temperature tends to increase by February/March and reaches the maximum in Summer. The maximum surface temperatures were 20°C (11th July 1977), 18.5°C (21st August 1978), 19°C (9th July 1979) and 20°C (4th August 1980). Decreasing and low temperatures coincided with Autumn - early winter periods and the coldest records (1.5 - 3°C) were made during mid-winter periods. Shearwater was covered by ice in the winter periods of 1978/1979 and during the latter period the overall coldest temperature (1.5°C) was recorded. The surface temperature data for

Shearwater indicate that the annual cycle resembles those of northern temperate lakes (HUTCHINSON, 1957).

The maxima of diatoms in the spring and early summer and of blue-green algae and green algae during summer and early autumn in Shearwater, suggests that temperature is one of the decisive factors in the growth of planktonic algae. Growth of representatives of the phytoplankton in Shearwater with respect to temperature will be considered in the section on seasonal periodicity of species.

Water level:

Changes in water level affect the nutrient balance of any water. The inflow and outflow rates influence the concentrations of dissolved substances in the water and alternatively its algal flora. Rise and fall of water level in Shearwater is shown in Fig. 2. The water level was usually low during summer of each year due to the loss by evaporation and slow inflows. However the water level starts to rise in September and continues to increase during autumn - early winter periods. There was usually an abrupt fall in the water level by mid-winter. The decrease appears to be sharper in 1978 and 1979 than that recorded in 1980, possibly owing to ice formation in the first two years. It is noteworthy that there was not a conspicuous increase in water level after the ice melted in Shearwater. There was also a rise in water level during Spring in Shearwater but on the whole water level in Spring was lower than that of Autumn - Winter period. The highest lake levels

Fig.2. Seasonal variations in lake level.


water level cm.
23
22
21
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19
18
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14
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J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J
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1978
1979
1980

were recorded during December in 1977/1978 and during March and February respectively in the two following years. The lowest levels coincided with July of 1977, 1978 and 1980 while in 1979 it was recorded in May. In lakes with a natural stream outflow the change in water level and the flushing out of populations can be a major though often neglected factor in phytoplankton ecology. In Shearwater, however, the existence of an overflow weir minimises the flushing.

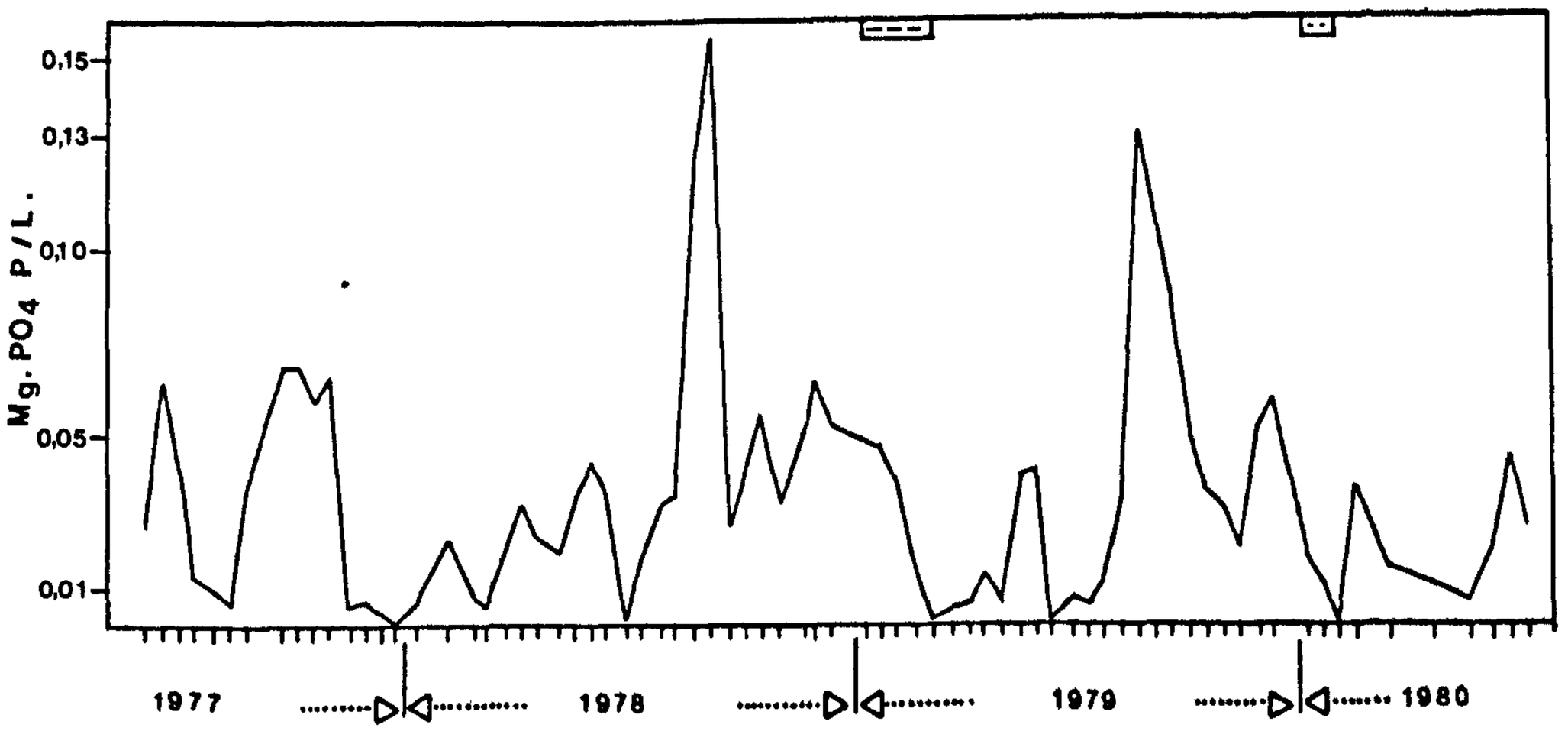
RESULTS OF CHEMICAL ANALYSES

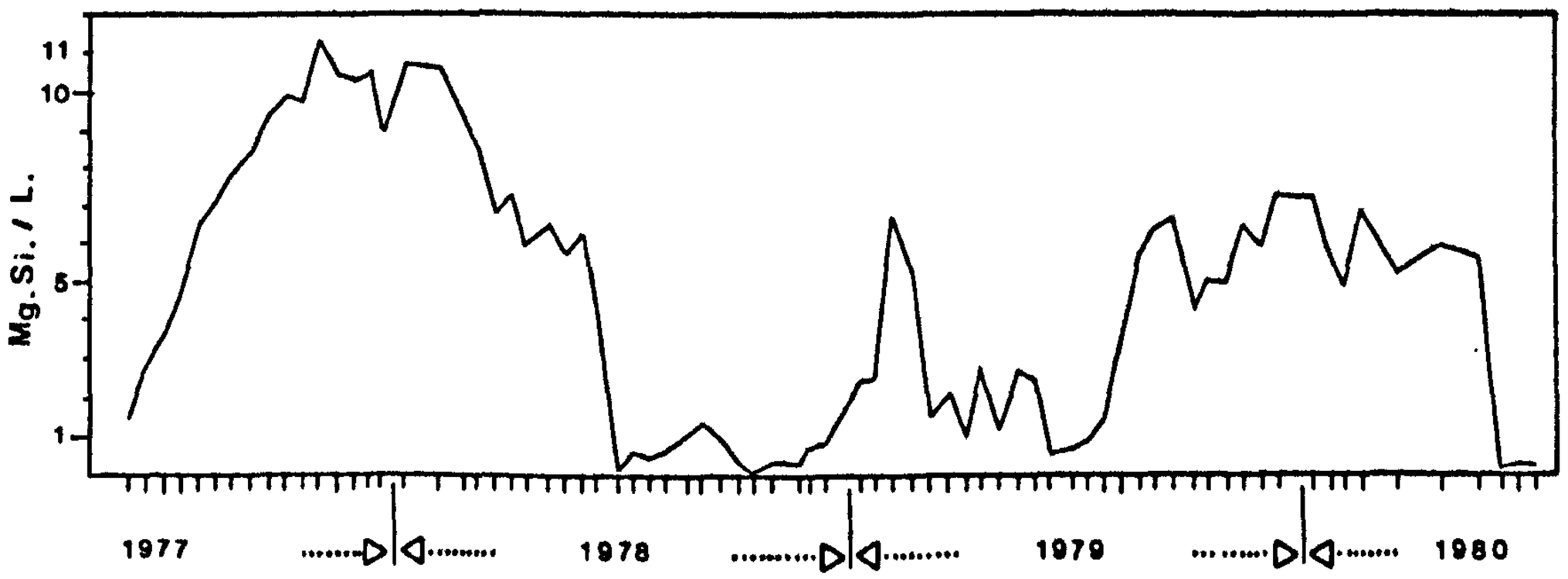
Nitrate nitrogen:

Nitrogen is one of the major constituents of cellular protoplasm of organisms and forms a major nutrient that affects the productivity in freshwaters.

The results of nitrate nitrogen estimations for Shearwater are shown in Fig. 3. It is apparent that the concentrations of nitrate in Shearwater exhibited a similar seasonal cycle in each year with little differences over the period studied. Concentration of nitrate tends to rise by the beginning of Autumn and reaches its Autumn peak within a short period. Sharp declines in concentrations regularly occurred after the Autumn maxima were achieved. Winter maxima commenced soon after Autumn peaks and comprised the higher levels of nitrate nitrogen. Maximum concentrations in the lake were recorded in March 1978 (1.25 mg./l) and in January (1.09 mg./l) and February (1.48 mg./l) in the two following years. A sharp decrease in nitrate was coincident with

Fig.3. Seasonal variations in concentration of phosphate (top), silica (middle) and nitrate (bottom).


0,15
0,13
0,10
0,05
0,01
Mg. PO4 P / L.
1977
1978
1979
1980


11
10
5
1
Mg.Si. / L.
1977
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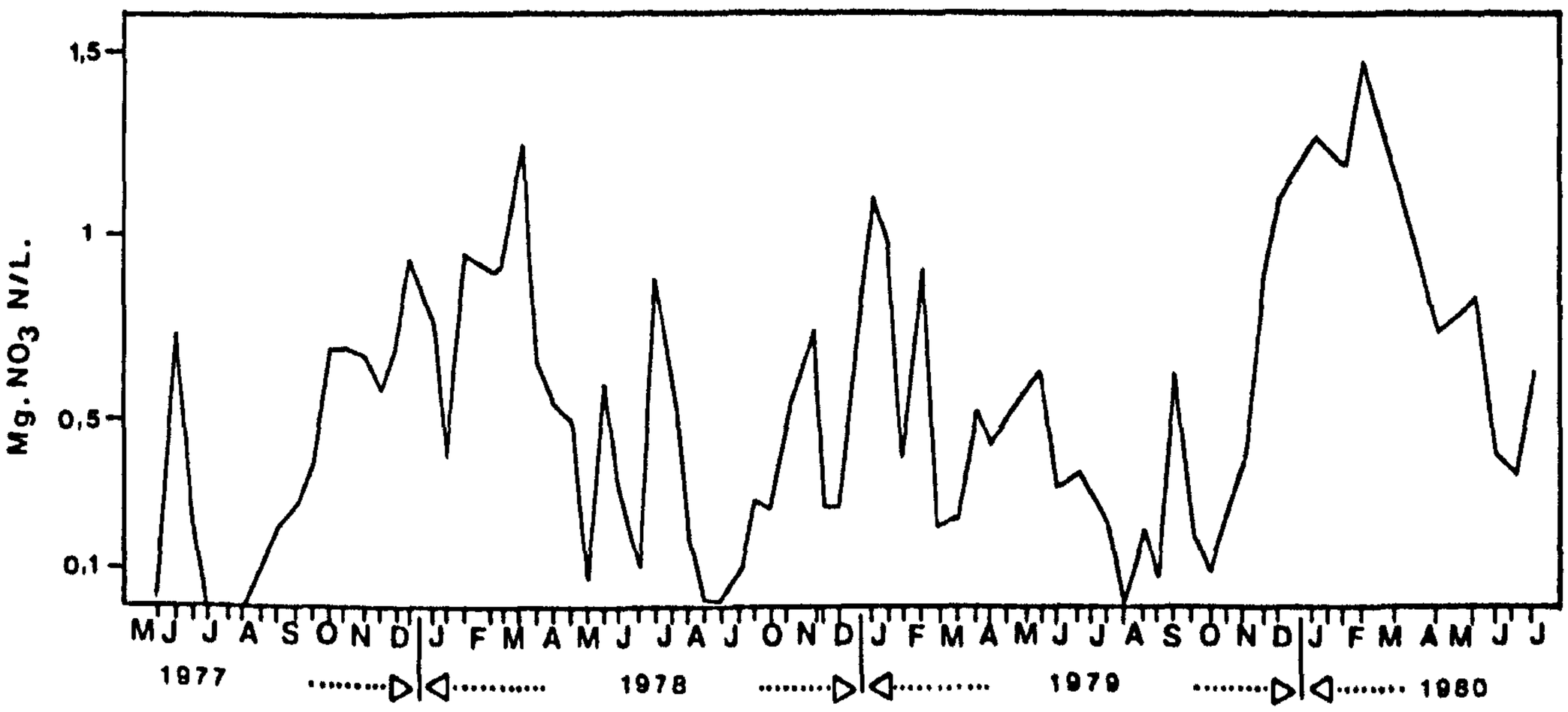
1980


1,5
1
0,5
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Mg. NO3 N/L.
M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J
1977
1978
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the ice formation in winter of 1978 whereas the reverse held true during the ice period in 1979 since nitrate reached its maximum level under the ice. A replenishment of nitrate in Winter (February) for a second time after the winter maximum (January) was also noted in 1979 although it usually decreased gradually after the winter maxima were achieved. Nitrate decreased gradually during the spring of 1978 and 1980 coinciding with the vernal decline of diatoms. However a replenishment of nitrate in the spring of 1979 appeared to synchronise with the active growth of diatoms particularly of Cyclotella spp. and Stephanodiscus hantzchii. March - April usually coincided with high levels of nitrate in Spring but by the end of Spring, nitrate was reduced again to low levels particularly in 1977 and 1978. The nitrate was replenished in Shearwater by early - mid summer in three years although a gradual decline was recorded all the way through summer in 1979. However, in general, summer periods were synchronous with the lowest concentrations in Shearwater.

The lowest concentrations and virtual absence of nitrate nitrogen occurred during July - August in 1977, August - September in 1978 and August in 1979 accompanying the blooms of blue-green algae (particularly Aphanizomenon flos-aquae).

Nitrate nitrogen content of the surface water in Shearwater with summer minima and winter maxima corresponds with increased utilization by phytoplankton and microbial activity. Similar patterns for nitrate nitrogen concentration have been recorded also for many freshwater lakes (see PEARSAL, 1930; LUND, 1956/1957 and HUTCHINSON, 1957). In addition, observations in winter, late summer and autumn and by LUND (1938) who found the maximum

to be in December and the minimum in August, are partly in agreement with the present study.

However, the present study does not substantiate the finding of WADE (1949/50) that nitrate concentration rises as the total phytoplankton decreases, since the phytoplankters in Shearwater were abundant at low as well as at high nitrate concentrations. A relationship between decrease in nitrate and increase in diatoms was suggested by FLINT (1949/50) which is in contrast to the present data since the diatom growth in Shearwater usually coincided with increasing concentrations of nitrate. However the present study supports, in part, the view of LUND (1956/57) that irregular relationship between phytoplankton crop and nitrate concentration often occur but fall in nitrate may continue as the crop declines.

ASMUND (1955a) noted irregular variations in nitrate concentrations and attributed the reduction in nitrate to lower pH which inhibits nitrification of humus substances. Irregular variations in nitrate were also characteristic in Shearwater, however the lowest concentrations were coincident with high pH levels. Nevertheless increase in nitrate was usually associated with rising water level during this study.

A remark of HERON (1961) that removal of nitrate in lakes commences after the depletion of SiO_4 is not in agreement with the present data since both nitrate and silicate decline simultaneously in Shearwater. However, during summer in 1978, a sharp rise in concentration of nitrate coincided with the depletion of silica during this investigation. LUND et al (1963) recorded the rise in nitrate during January - early May,

subsequent rapid fall after May and its replenishment in November. However, the declining or low concentrations of nitrate usually occurred in May and its replenishment after May appeared to be regular with autumn replenishment starting as early as September.

Phosphate:

Phosphate phosphorus has a major role in biological metabolism and most commonly limits the primary productivity in waters.

The phosphate (PO_4-P) cycle was irregular in Shearwater (Fig. 3.) but it can be summarised in the following way. The periods late summer - autumn were associated with the highest levels of phosphate in Shearwater (highest concentrations being in September 1978 (0.15 mg./l) and August 1979 (0.13 mg./l)). Concentrations of phosphate were either low or decreasing during winter and spring accompanied by large diatom crops. However some irregular peaks as well as relatively reduced concentrations were also recorded during winter - spring periods. An increase in phosphate was followed by a decline under the ice in 1978 while phosphate was gradually decreasing in 1979 when Shearwater was frozen over again.

Concentrations of phosphate were reduced to very low levels or actually exhausted by early summer but replenishment commenced by mid-summer usually proceeding to maximum levels either in August or early autumn. The lowest concentrations or exhaustion of phosphate were noted in Decerber 1977, March and June 1978/79 and in February 1980.

PEARSALL (1930) found phosphate at high levels in winter or early spring and minimum levels during the summer while RAO (1955) recorded high concentration in winter, late summer - autumn. Both observations, in part, are in harmony with the annual cycle of phosphate in Shearwater.

ATKINS (1926a) and HARVEY et al. (1935) correlated the decrease in phosphate with increase of diatoms and the first author associated the variations in phosphate with light rather than temperature. REYNOLDS (1973) also found that the concentration of phosphate phosphorus was frequently reduced in the periods of active diatom growth. Concentrations of phosphate in Shearwater were also often reduced to low levels during the active growth of diatoms. However increasing concentrations of phosphate also coincided with the diatom increase in Shearwater and at one stage the overall highest concentration was synchronous with active multiplication of diatoms particularly Cyclotella spp. and Stephanodiscus hantzchii. Hence the concentration of phosphate did not appear to be influenced by diatom growth in this study supporting the view of LUND (1956/57). The present study is also in accord with HAMMER (1964) who found considerable increases of phosphate phosphorus on the death of blooms of blue-green algae. There were always sharp increases in phosphate level after the termination of blue-green algal blooms particularly of Aphanizomenon flos-aquae in Shearwater.

Excretion of phosphorus by zooplankton, particularly as phosphate,may influence competition between planktonic algae and their spatial distribution. COOPER (1935) recorded the

abundance of zooplankton being associated with increasing concentration of phosphate. Although the number of zooplankters in Shearwater remained low, increase in the numbers of zooplanktonic species usually coincided with rising phosphate. COOPER (1935) and HUGHES & LUND (1962) stated that the phosphate is also utilized by bacteria which may affects its concentration rather than the increase in diatom populations.

STRICKLAND (1960) suggests that the rate of absorbtion of phosphorus by phytoplankton depends upon the ratio of phosphorus to nitrogen. HERON (1961) and SWALE (1964) correlated high phosphate levels with rainfall and water level. Phosphate decreased or rose in Shearwater without showing a clear relationship with rising or decreasing water levels. In addition on one occasion depletion of phosphate coincided with the overall highest water level during this study. Influence of surface water temperature on fluctuations in concentrations of phosphate was also uncertain in Shearwater. Low concentrations were recorded in the periods of very cold as well as warm weather and sharp peaks occurred regardless of the temperature. However two distinct maxima of phosphate were achieved during warm periods.

Silicate:

Diatoms form a major part of the phytoplankton in many lakes and over large areas of the oceans and their development is always associated with adequate silica levels.

The seasonal cycle of dissolved silica in Shearwater showed more or less a similar pattern in each year but with variations

in concentration over the period studied (Fig. 3.). Winter periods coincided with the richest concentrations of dissolved silica while lower concentrations were recorded in autumn and spring. However the concentration of silica remained more or less constant during autumn and spring, as there was little utilization. Silica appeared to be lowest in concentration during summer except for 1978 when the autumn was lower than summer. In general, in 1977 - 1978 and 1979 - 1980 concentrations of silica were higher than 1978 - 1979 if the June to June cycle of silica is considered. It is of interest to note that rapid falls and equal replenishments of silica were characteristic of the annual cycle of this dissolved nutrient in Shearwater.

Concentration of silica tends to rise in summer and continues to increase through autumn and winter even accompanying the active growth of diatoms. Silica starts to decline by late winter and decrease continues through spring, indicating the large amount of utilization of silica by the spring development of diatoms (e.g. Asterionella formosa). By early summer (usually June) concentration is either reduced to very low levels or actually exhausted in Shearwater. The highest concentrations were recorded in October 1977, February 1979 and December - January 1979 and 1980 while the lowest levels occurred in June during this investigation. However exhaustion of silica was also noted in October 1978 although such periods usually coincided with increasing and in one case with the overall highest concentration of silica recorded in Shearwater. Ice formation did not appear to influence the concentration of silica. A slight decline of silica was synchronous with ice period in 1978 during which the diatoms (particularly Asterionella) were also increasing actively

thus masking the effect of ice. In addition, during the icy winter of 1979 silica reached its maximum level under the ice.

The seasonal cycle of silica in Shearwater is in harmony with the observations of PEARSALL (1930) who also reported high concentrations in winter or early spring and minimum silicate during the summer. Present data also agrees, in part, with those of MORTIMER (1941), FLINT (1949/50) and RAO (1955) who noted the high silica content in summer, autumn and winter. MORTIMER (1941) attributes the summer rise to production in the hypolimnion and also to an increase in water level and solution from sediments. Although there was an increase in silica content coinciding with the rising water level in Shearwater, silica fell or rose regardless of the variation in water level. FLINT (1949/50) recorded a minimum level of silica in spring, a finding not in accord with the present study. ASMUND (1955a) also reported high silicate in January - February and low concentrations from March onwards, a similar cycle of silica to that recorded during this investigation. However the present data differed from that of HERON (1961) who found that silica rose steadily from May to August and then remained more or less steady until October.

PEARSALL (1930) and SWALE (1964) suggested that the fall in silica content coincides with the vernal diatom maxima, a finding not substantiated by the present data. Vernal developments of diatoms, particularly Asterionella,were synchronous with constantly increasing concentrations of silica in Shearwater and increase in silica continued until the vernal maxima were achieved. Diatoms were quite abundant in Shearwater (during late winter - spring periods) when concentrations of silica were relatively high.

Moreover silica content was usually decreasing simultaneously with the vernal decline of the diatoms, this also conflicting with the study of GARDINER (1941a) who recorded an instant rise in silica with the decline of the vernal diatom phase. Long term studies on Asterionella showed that the decrease in numbers of this diatom has coincided, over a period of many years (LUND, 1950; LUND et al., 1963) with a drop in dissolved silica concentration below 0.5 mg/l. This agrees with an observation of PEARSALL (1932) that large maxima of diatoms will not arise if the concentration of silica is below this level. JØRGENSEN (1957), however, found the development of various freshwater diatoms both in culture and in lakes to be limited at somewhat lower concentrations (0.03 - 0.04 mg./l) than 0.5 mg./l. Concentrations of dissolved silica in Shearwater dropped below 0.5 mg./l. on a few occasions, two of which coincided with the achievements of maxima of Cyclotella and Stephanodiscus (October 1978) and of Fragilaria (June 1978). This shows the large utilization of silica by diatoms. Active growth of Fragilaria in Shewarwater during low levels of dissolved silica in early summer and vernal development of Asterionella with high levels of silica suggest that the different diatom species have quite varied silica requirements. Attempts to correlate growth of individual diatom species with variations in silica will be made in a later section.

CHAPTER 2.

PHYTOPLANKTON OF SHEARWATER

Shearwater is an eutrophic body of water supporting large crops of phytoplankton which has not been investigated previously before this study. However each year phytoplankters undergo a more or less similar pattern of growth in Shearwater with autumn - early spring dominant diatoms, followed by active growth of blue-green and green algae during late spring - summer.

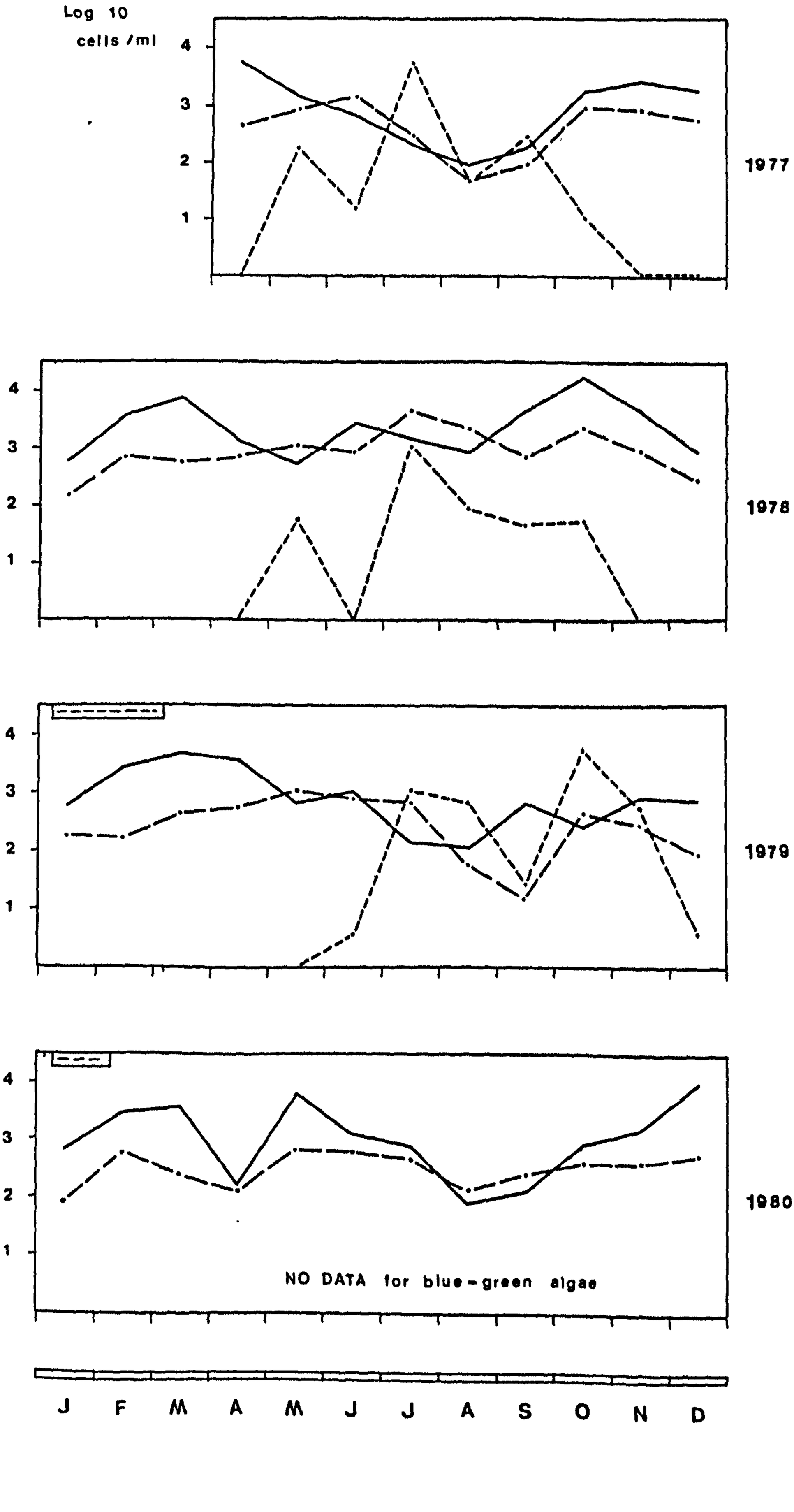
Total numbers and the annual cycle of blue-green algae, diatoms and green algae are shown in Fig.4. It is apparent from the same figure that diatoms and green algae were present in Shearwater at all times of the year while blue-green algae appeared only in the second half of the year. Fluctuations in the total numbers of diatoms and green algae were more or less simultaneous while blue-green algae were most abundant in the periods of declining or low numbers of diatoms and green algae. In general the diatoms were the most abundant group followed by green and blue-green algae respectively. However, domination of these groups of algae differed in certain periods of the year. The rapid rate of active growth of the dominant phytoplankters was maintained until the environmental conditions altered or some factor became limiting. Hence the composition of the algal standing crop is related to the physico-chemical and biological factors. Some members of the phytoplankton were growing rapidly and having large maxima (e.g. Asterionella,

Fig.4. Annual cycle of the three major populations

---- blue-green algae (Cyanophyceae)

—— Diatoms (Bacillariophyceae)

-·-· green algae (Chlorophyceae)


Log 10
cells /ml
4
3
2
1
1977
1978
1979
1980
NO DATA for blue-green algae
J
F
M
A
M
J
J
A
S
O
N
D

Pediastrum, Aphanizomenon) while some merely persisted in the surface layer, but did not multiply at sufficiently rapid rates to produce dominant populations in the phytoplankton, e.g. Melosira spp., Staurastrum. However, a few of these less numerous organisms remained in the phytoplankton for a long time, indicating that they have a wide tolerance to changing environmental conditions. However these scarcer planktonic species require a narrower ecological niche (HUTCHINSON, 1967) to enable them to grow exponentially, and thus they are only able to maintain themselves in the plankton at a slow rate of growth.

It is obvious that specific environmental conditions are required by other organisms to reach to optimum growth rate. The dominant planktonic algae reflected the conditions in Shearwater since only a few species reappear in large numbers in each year. Large blooms of Asterionella regularly occurred each year in late winter - spring during which similar physico-chemical conditions predominate. Again in summer similar conditions coincided with the blooms of Aphanizomenon (blue-green alga), Coelastrum, Pandorina and Pediastrum (green algae). However water conditions obviously differed from year to year and the planktonic algae causing blooms in Shearwater occurred in different proportions in each year, clearly demonstrating slightly differing environmental conditions.

Seasonal Cycles of Planktonic Algae in Shearwater

The detailed seasonal variation in cell numbers (colonies and coenobia in the case of a few Chlorococcales and blue-green algae) is obivous from the graphs of individual species. These variations will not be described in written detail but any particular notable features will be commented upon and general seasonal variations and correlations between species and environmental factors will be highlighted.

BACILLARIOPHYCEAE

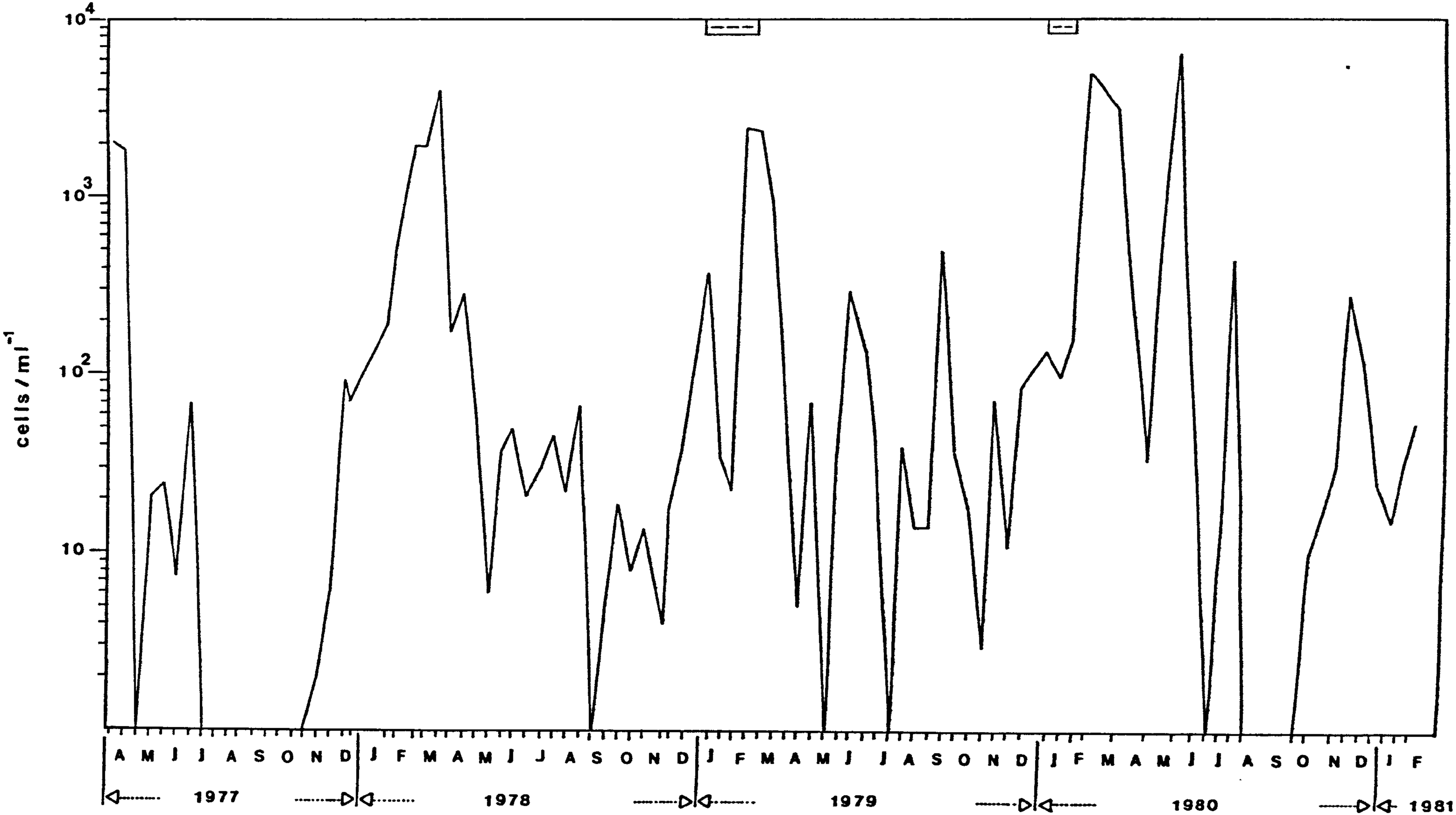
The members of the Bacillariophyceae constitute a large part of the phytoplankton in Shearwater. Three species of diatom were responsible for the blooms over the period studied. They were Asterionella formosa, Cyclotella, sometimes mixed with Stephanodiscus and Fragilaria crotonensis.

Asterionella formosa Hass.

Asterionella formosa is a conspicuous member of the phytoplankton in Shearwater and exhibits a definite seasonal variation (Fig. 5.).

During this study Asterionella was present in the plankton of Shearwater on most occasions. The periods of low numbers or virtual absence coincided with the mid-summer/early autumn periods especially in 1977 and in 1980. In the other two years

Fig.5. Seasonal periodicity of Asterionella formosa


10^4
10^3
10^2
10
cells / ml^{-1}
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F
1977
1978
1979
1980
1981

Asterionella was more abundant and the lowest counts were recorded in September, May and July. However, summer periods are also known for the low count of Asterionella in many freshwaters (LUND, 1949; KARIM, 1964; HAPPEY-WOOD, 1968; HICKMAN, 1970).

The growth of Asterionella in Shearwater, however, displayed some different features than those recorded in Windermere (LUND, 1949, 1950) and Abbot's pond (KARIM, 1964; HAPPEY-WOOD, 1968; HICKMAN, 1970). Growth of Asterionella in Shearwater may be compared with physico-chemical factors, shown in Fig.5 and Fig.3.

To begin with, it is apparent from the Fig. 5. that the onset of the spring maximum tends to occur in October/ November in Shearwater while the temperature and illumination are decreasing. Thus supporting in part WHITFORD's (1960) view that Asterionella has a low temperature and high light requirements but in disagreement with LUND (1949) that reduced water temperatures reduce the rate of growth.

In addition, CANTER & LUND (1951) reported that the gradual increase of Asterionella starts as the temperature increases; whereas in Shearwater, in three years the gradual increase to the vernal maxima synchronized with the temperature decrease. Only the vernal development in 1980 appears to be supporting the view of CANTER & LUND since it commenced when the temperature started increasing.

The commencement of vernal maximum being as early as October/November in Shearwater differs from the situation in most lakes, see the studies of LUND (1949), KARIM (1964),

HAPPEY-WOOD (1968) and HICKMAN (1970) who recorded the onset of the vernal growth mostly in January/February, occasionally as late as March, when the temperature and illumination start increasing. However adaption of communities to low light intensities has been demonstrated (STEEMAN & HANSEN, 1959; TALLING, 1966) although it was stated that incipient light levels may be the most important factor in determining the onset and maintenance of growth (RODHE, 1948; LUND, 1949; TALLING, 1960, 1971).

Moreover, at one stage (1979), vernal maximum of the alga was recorded when Shearwater was covered by ice.

Ice formation did not affect the development of Asterionella when Shearwater was covered by ice in 1979 and in 1980. Although there was a slight decrease in the numbers of the alga, this decrease was masked by the parasitism of Z. affluens Canter on the population. This finding is in accord, in part, with HUTCHINSON (1944) who found Asterionella in Linsley pond reached a maximum under the ice and during the spring it was rare and sporadic, but in disagreement with LUND (1959) and KARIM(1964) who reported that the ice cover affected the development of Asterionella which disappeared from the samples at such times. However, this effect may be due to the degree of turbulence remaining in the water column at the period when ice forms.

Vernal maxima in Shearwater mostly occurred in February and March at very low temperatures around 5 - 7^{o}C and at low light intensities whereas in many freshwater lakes the initial onset of the vernal growths tend to occur in March and the

maximum is generally reached during the warmer and high illumination period of the year (LUND, 1949; KARIM (1964). The onset and the actual occurrence of vernal maximum at low temperatures and light intensities is also in disagreement in part with REYNOLDS (1973) who reported that, of all the environmental factors, light level appears to be critical in determining the onset of growth and the size of population. The size of the last vernal maximum of Asterionella in Shearwater was the greatest and it occurred in the period of the warmer and the higher illumination in contrast to previous years. In 1980, an unusual situation occurred in Shearwater when 3183 cells/ml were recorded in March, numbers declined to an unusual 33 cells/ml after a fortnight due to unknown factors, and this was followed by an equally unexpected growth to the spring maximum of 7309 cells/ml two weeks later. This situation disagrees with LUND (1950) who stated that the density of the population rises until all the available silica has been used in the second half of April, when the supply of silica is replenished from the inflows growth may be renewed for a short time but no large population is produced. In addition, vernal maxima of Asterionella in Shearwater occurred always when the concentration of silica was quite high - over 5 mg/L - showing that the increase of the population does not continue until all the available silica is used by the alga since the numbers of Asterionella otherwise should have continued to increase using up the available silica in the water. Thus the present study would suggest that the increase in the numbers of Asterionella is controlled by other factors as well as the amount of silica in the water.

LUND (1950b) stated that the spring maximum ends at the time of maximum illumination and higher temperatures but that these two factors are not responsible for the decline which is frequently due to depletion of the available silica. In the present study the vernal maxima of Asterionella generally ended much before the maximum illumination and higher temperatures but on two occasions depletion of silica was also noted (apart from 1980). Thus supporting the view of LUND (1950b) only in that high light intensities and temperatures are not responsible for the termination of vernal maximum but may be the depletion of silica.

Two maxima were reported by WEST & WEST (1912) for Asterionella and LUND (1949) considered that the spring maximum is far greater than the autumn maximum. Although there were no autumn peaks in Shearwater in 1977/1978, they did however occur in September and in November in the other two years. They were much smaller than those of the spring maxima, thus supporting the suggestion of LUND (1949).

In addition to the spring and autumn maxima, there were also regular summer peaks in Shearwater which occurred in August 1978 and in June in the other three years. The occurrence of summer peaks coincided with the complete end of spring maxima and high temperature and illumination. Summer maxima in 1978 and in 1980 were greater than autumn maxima although this was reversed in 1979.

RUTTNER (1953) and HUTCHINSON (1967) designated plankton organisms as oligothermal (cold-requiring) polythermal (warmth-requiring) or eurythermal (tolerant to wide temperature range). Asterionella in Shearwater appeared to be eurythermal considering its growth in three different seasons.

Summer peaks were also recorded in other freshwater bodies (LUND, 1949; KARIM, 1964; HICKMAN, 1970 and REYNOLDS, 1972) occurring just after the spring maximum ends in June/July. In addition in some cases the summer peaks were also greater than the autumn maxima (as was found in Shearwater). In addition, WESENBERG-LUND (1908) stated that diatoms are not necessarily inhibited by higher water temperatures.

The occurrence of summer peaks in Shearwater was more regular and in most cases greater than the autumn maxima. Considering all these regular features of the summer peaks in Shearwater and also its occurrence in the other freshwater bodies, the present study would suggest that it is possible for three maxima to occur in any one year if the conditions are favourable.

The period of Asterionella growth lasted more than 10 weeks (usually 16 weeks in Shearwater) although GARDINER (1941a) observed that the development of Asterionella is restricted to a few weeks due to complete or partial exhaustion of plant nutrients other than silica and phosphorus. LUND (1961) also refers to this unknown nutrient or nutrients which affect the development of Asterionella.

The present study is not in accordance with the conclusion reached by LUND et al. (1963) that the wetter and the colder the winter, the smaller the size of the subsequent Asterionella crop. During the present study, the average winter temperatures of each year were very similar and very cold but the size of the standing crop varied considerably.

The onset of the vernal maximum of Asterionella showed a marked relationship with nitrate and silica in Shearwater since each spring maximum started with either increasing or high concentrations of nitrate and silica. However the correlation between the phosphate and the onset of vernal maximum did not appear to be as marked as of nitrate and silica. Although the concentration of phosphate was the lowest in 1977, in the two following years the onset of the vernal maximum coincided with increasing phosphate concentrations. Therefore the present study generally supports PEARSALL's (1932) view that Asterionella occurs when nitrate, phosphate and silicate are abundant.

The phosphate did not show a clear correlation with the vernal increase of Asterionella either in Shearwater. The concentration of phosphate was either decreasing or increasing during this period.(Fig.3.). However, generally speaking the phosphate level was declining after the first step of the vernal development and by the time the vernal maxima was reached, the concentration was reduced to very low levels indeed. In addition, during the vernal decline of the alga the concentration of phosphate was always rising in Shearwater.

During autumn and summer development of Asterionella, the concentration of phosphate was usually increasing, although a decrease was recorded in 1977. The maxima of autumn and summer developments usually coincided with high phosphate levels but at one stage a summer maximum (June, 1979) was also reached when the phosphate was at its lowest level.

Thus the present data substantiates RODHE (1948) who

reported that phosphate dependency of the growth of Asterionella is of a very complicated nature and phosphate utilization is, to a high degree, affected by one or more unknown factors in the lake water. MACKERETH (1953) also found little correlation between growth of Asterionella and change in phosphate concentration in Windermere. However LUND (1950) and LEHMAN (1979) showed the ability of Asterionella to store excess quantities of phosphate in culture. Therefore this might be the reason for the increase or small peaks of Asterionella at very low levels of phosphate in Shearwater.

Although there were some fluctuations in the concentration of nitrate during the vernal increase of Asterionella, the general pattern of nitrate was of rising values during these periods in Shearwater. In addition nitrate was almost at its highest levels when the vernal maximum occurred in 1978 and 1979. In 1980 nitrate was actually at maximum level when the first vernal peak of Asterionella occurred, and then started decreasing gradually. However by the time the vernal maximum of Asterionella ended, the concentrations of nitrate were very low indeed in Shearwater. Nitrate always returned to the water after the vernal maxima finished.

During the 1977 - 1978 vernal increase of Asterionella, silica was declining simultaneously and the decline continued after the vernal maximum ended. In 1979, however, silica concentration continued to increase alongside the vernal Asterionella maximum. In 1980, during the vernal increase, it was rising again but exhausted by the end of the vernal maximum.

Thus the present data disagrees with LUND (1950a) who found that the concentration of nitrate usually declines and silica always does during the development of an Asterionella maximum.

Summer and the autumn appearances of the alga coincided with rising nitrate and silica levels. The concentration of nitrate always decreased while silica either decreased or continued to increase after the maximum peaks of summer and autumn developments.

In conclusion, the present data would suggest that the occurrence of Asterionella coincides with high concentrations of nitrate and silica and these nutrients support the vernal development of Asterionella almost until the actual spring maximum is reached.

LUND et al (1963) and TILLMAN et al (1976) emphasized silica and phosphate as major factors governing the abundance of Asterionella and LAZERTE (1980) suggested that silica is the dominant factor limiting standing crops of Asterionella. In the present study during 1978 - 1979 vernal development of Asterionella the total concentration of phosphate was higher than those of other years but the size of standing crop was the smallest of all maxima recorded. In addition, the silica level during 1977 - 1978 spring increase was much higher than that recorded during 1979 - 1980 but the vernal size of the latter was almost double the size of the former. Thus the present study would suggest that the abundance of Asterionella depends on other factors as well as the concentrations of phosphate, silica and possibly nitrate. The size of the vernal

maximum of the alga showed a clearer correlation with the concentration of nitrate in Shearwater as the lower concentration of nitrate coincided with the smaller size and the higher concentration with the greater size of the standing crop of Asterionella formosa.

In 1977, Asterionella was absent from the plankton samples for more than three months (July - November) (Fig although the concentrations of nitrate, phosphate and silica were increasing rapidly and/or at high levels. There was no chytrid infection during this period either. Grazing would not account for this absence as LUND (1961) states that no animal is known grazing extensively on Asterionella. Therefore the factors which are responsible for the absence of Asterionella remain obscure, supporting the view of CANTER & LUND (1951) that some factor other than silica supply limits the growth of Asterionella and usually other diatoms sometime after thermal stratification sets in, usually June or July, so that growth is not renewed until September or October.

Variation in number of cells per colony

The colonies of Asterionella in Shearwater consisted predominantly of 4 cells although 8 celled colonies were abundant during 1980. Analysis of cell numbers of colonies yielded the following data:-

Cells/colony	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
January-December 1978	217	298	198	577	66	43	46	99	2	2	3			1		1
January-December 1979	573	377	173	1220	58	28	5	42		6		2			6	
January-December 1980	346	350	118	772	56	109	120	500	16	18	6	10	8	22	36	69

Table I. Annual total number of colonies with different cell numbers.

It is obvious from this table that numbers higher than 8 cells per colony were not common. The dominant number of cells/colony is 4 particularly if the lower numbers are considered to be associated with 4 celled-colonies. However 8 celled-colonies were also considerable in 1980. (Table I.). WEST & WEST (1905) also reported the common 8 celled-colonies. The persistance of 4 celled-colonies is in harmony with the study of GRIFFITHS (1925).

Seasonal distribution of colonies with different cell numbers is shown in Fig. 6. This diagram shows that single Asterionella cells are always present when the alga is. The number of cells per colony starts increasing with the vernal development of Asterionella.

The number of 8 celled-colonies rises at the start of the vernal increase of Asterionella, and continues to increase along with the vernal development. The actual vernal maximum of Asterionella coincides with the highest numbers of 8 celled-colonies. 16 celled-colonies were encountered only at the time when the vernal maxima of Asterionella occurred. Fig.6. also shows that the greater the size of the vernal maximum the higher the numbers of 8 - 16 celled-colonies. Nevertheless the 4 celled-colonies were more common than 8 - 16 celled-colonies during the spring development of Asterionella in Shearwater. In addition, it is interesting that a vernal maximum of Asterionella consisted mainly of 4 celled-colonies although there were some 8 celled-colonies.

In the summer periods,single cells and colonies with 2 - 3 cells were more common, particularly single cells, due

Fig.6. Seasonal distribution of the colonies of different cell number of Asterionella formosa.

Upper section: _____ 1 celled colonies

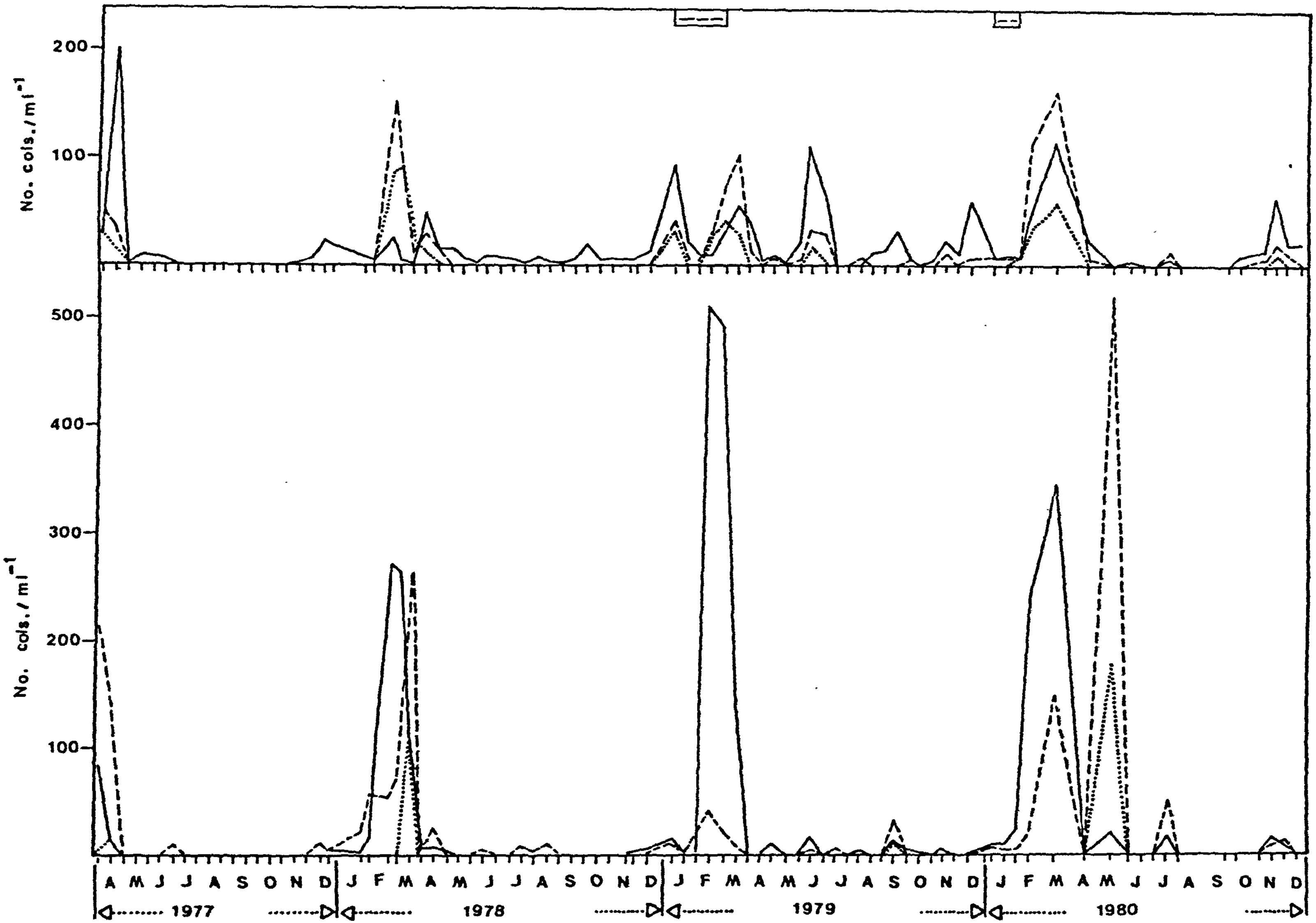
_ _ _ _ _ 2 celled colonies

..... 3 celled colonies

Lower section: _____ 4 celled colonies

_ _ _ _ _ 8 celled colonies

..... 16 celled colonies


No. cols./ml⁻¹
200
100
500
400
300
200
100
No. cols./ml⁻¹
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

to less active multiplication of the alga under less favourable conditions compared with the period of spring. However a summer peak (1980) was formed mainly by 4 - 8 celled colonies.

In conclusion the number of cells per Asterionella colony appeared to be related to the rate of active multiplication of the alga.

The present study is in disagreement with GARDINER (1940) who found that 8 - 16 celled colonies were abundant at the start of the period of Asterionella growth, then they decreased later in the same period of the growth. In the present data 8 - 16 colonies were few at the start but reached their highest number with the vernal maxima of Asterionella. These studies agree with LUND (1959) who found that the number of cells per colony was 8 - 16 during active multiplication and they decreased when the environmental conditions became unfavourable.

LAZARTE (1980) also found that Asterionella colonies were usually 1, 2 and 4 celled and around May 7 - 8 celled colonies dominated briefly. However TILMAN et al.(1979) found that the number of cells per colony increases (up to 20 cells per colony) in silica limited laboratory populations whereas under phosphorus limitation the number of cells per colony was reduced (from 8 down to 2 cells per colony).

Summary and conclusions

Asterionella formosa was a conspicuous member of phyto-

plankton in Shearwater with a distinct seasonal cycle and was present on most occasions.

Gradual vernal development of the diatom commences usually as early as in October/November when the temperature and illumination is decreasing.

The coldest and the lowest illuminated periods are quite favourable for the growth of this alga since vernal maxima usually occurred during such periods.

Ice formation does not affect the development of Asterionella: on the contrary, a maximum was achieved under ice.

The end of the vernal maxima does not show a correlation with maximum illumination and high temperatures. Vernal maximum usually terminates before such a period occurs.

Apart from regular late winter - spring maxima, regular summer peaks are very characteristic while autumn maxima are sporadic in occurrence. Hence three maxima occur for Asterionella in Shearwater. Summer maxima were usually larger than thoseof autumn.

Asterionella appears to be tolerant to a wide temperature range considering the maxima during low and high temperatures.

Dissolved silica and nitrate support the growth of the diatom until the vernal maxima are achieved. The declines in concentrations of nitrate and silica commences after this stage. The influence of phosphate on the growth of Asterionella appears to be complicated.

The size of the vernal maximum does not show a correlation with amount of available phosphate or silica but with nitrate.

Absence of Asterionella during the period of increasing concentrations of nitrate, phosphate and silicate suggests that some other factors are also governing the development of the species as well as dissolved nutrients.

The period of Asterionella growth during the vernal maximum lasts approximately 16 weeks.

The number of cells per colony increases with the vernal development of Asterionella and reaches highest numbers at the vernal maximum.

Fragilaria crotonensis Kitton

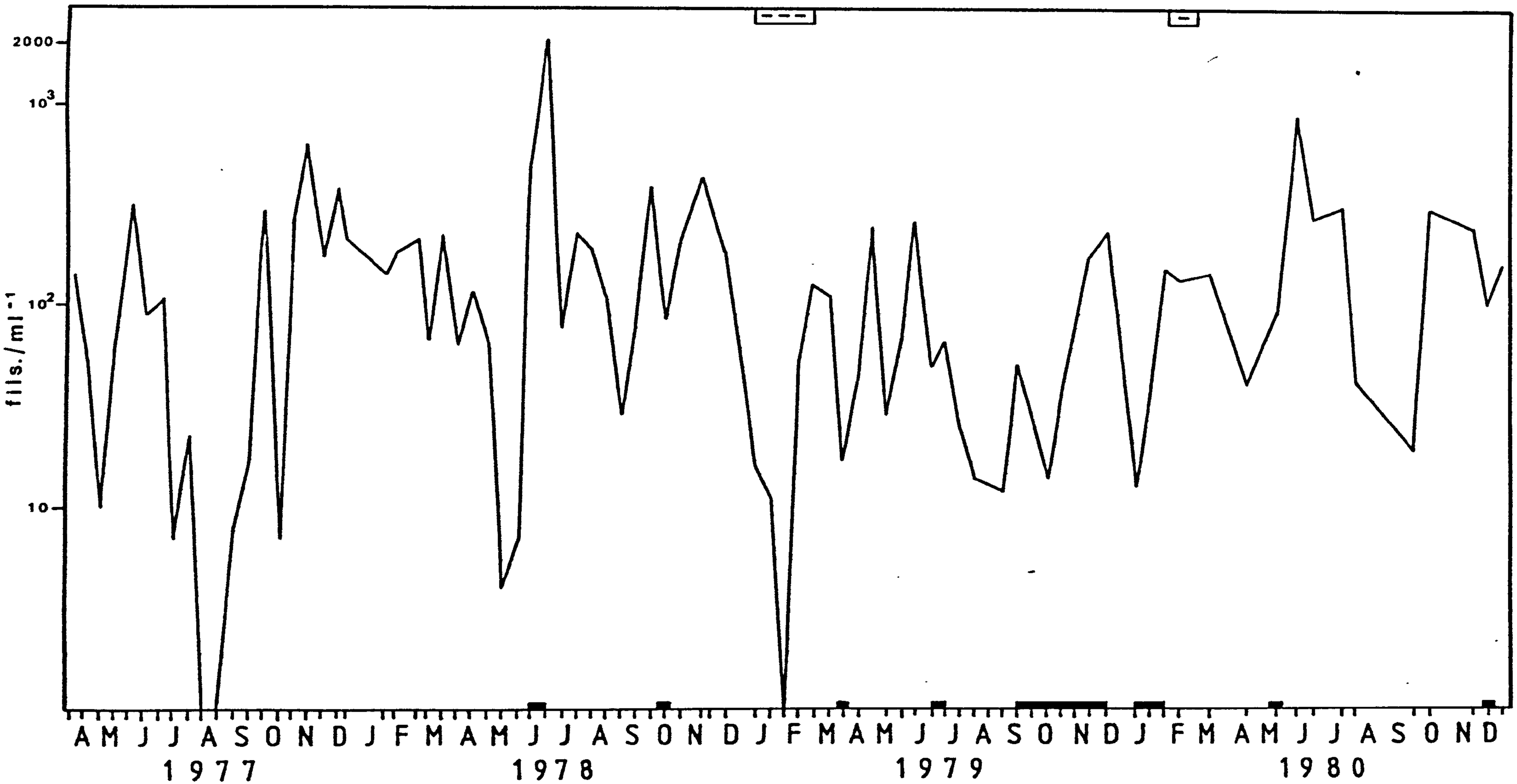
F. crotonensis was present for a longer time than A. formosa in the phytoplankton of Shearwater during this investigation. The seasonal fluctuations in the numbers of filaments of Fragilaria for nearly four years is shown in fig. 7.

Fragilaria was more abundant in 1978 and 1980 than in the other two years and was virtually absent in only three samples over the period studied. Early summer and autumn maxima of the diatom were quite conspicuous, indicating the main periods of active growth for Fragilaria. In June 1978 and 1980 the size of the summer maxima were 3 - 4 times as large as those of the autumn whilst in 1979 they were very alike in size. Fragilaria was generally in smaller numbers during winter and spring and it was replaced by blue-green and green algae after its summer maxima in late summer (July - August). Its seasonal cycles, therefore, are similar to those recorded in many north temperate lakes (e.g. WESENBERG-LUND, 1908). F. crotonensis was also found as an abundant species in an oligotrophic lake (RUTTNER, 1959) as well as in eutrophic lakes (HUTCHINSON, 1967, p.387).

Autumn growth of Fragilaria in each year tends to commence sometime in September and the maximum was achieved within 2 - 3 months. In 1977 and 1978 the autumn maximum was recorded in November and in two following years it occurred in December and October respectively. In the first two years the sizes of the autumn maxima were larger than those of subsequent years.

Fig.7. Seasonal periodicity of Fragilaria crotonensis.

▬▬ periods of fungal infection


2000
10^3
10^2
10
fils./ml^-1
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

In 1977 and 1978 a conspicuous growth peak of the diatom occurred in October before the actual autumn maximum was achieved. These peaks were recorded soon after the autumn multiplication of Fragilaria commenced, showing how fast the diatom can multiply within a short time when the conditions are favourable. In 1977, the size of the October peak was smaller than that of the autumn maximum while the sizes were very much similar in 1978. It is worth mentioning that there was a sharp decrease in the numbers of the diatom between the peaks (in October) and the autumn maxima (in November), coinciding with the decreases in the concentration of silica in both years (Fig. 3.).

The onset of the summer maxima of the diatom was considered as May in 1977 and 1978, and April in the two subsequent years. Summer maxima occurred within a shorter time (1 - 2 months) than autumn maxima and the rate of active multiplication also appeared to be much faster than during the autumn increase. Summer maxima in 1978, 1979 and 1980 all occurred in June and in the incompletely sampled 1977 it was recorded at the end of May. The summer maximum in 1978 was the largest (2149 fils./ml.) of all. In 1979, however, two peaks of growth occurred, one in April and one in June, and both were approximately the same size (circa 300 fils./ml.). This kind of growth pattern appeared to be a characteristic feature of the diatom since similar sharp peaks and declinations also occurred before the autumn maxima of Fragilaria were achieved in the first two years.

During winter and early spring, the numbers of filaments

were low compared with those of autumn and summer although a few sharp growth peaks also occurred during winter - early spring periods.

The lowest counts and virtual absence of F. crotonensis were recorded in mid-summer/early autumn period in 1977, middle of May in 1978 and during winter in the two following years.

Therefore, the seasonal periodicity of F. crotonensis in the present study, is in accord with WESENBERG-LUND (1904) who also found the species to be an abundant component of the late spring and of the autumn period in some lakes of Denmark. LUND (1964) also reported good growth of F. crotonensis in late spring/summer and autumn crops in the northern basin of Windermere. The present data also agrees with ALPSTEIN (1896) who found summer maximum of Fragilaria occurring in June, which is in conformity with the present study. However, the seasonal cycle of the diatom in Shearwater differs from those observed in the studies of RILEY (1940) and HUTCHINSON (1944) who reported that F. crotonensis began to increase shortly after July and became dominant in August in Linsley Pond.

Physical-chemical data for Shearwater are shown in figs. 2 & 3 and may be compared to the seasonal periodicity of Fragilaria (Fig. 7.).

Autumn increase of the diatom was synchronous with rising concentrations of silica in 1977 and 1979 while the concentration was decreasing in 1978. The onset of the autumn growth always coincided with a rising nitrate level and the phosphate supply was rich enough. Autumn maxima of Fragilaria was achieved when silica was at a very high level in 1977 and 1979 and at a very low level in 1978. Growth peaks in autumn (October) 1977 - 1978 also occurred at quite high concentration of silica.

The concentrations of nitrate, phosphate and silicate were increasing by the onset of the summer maxima of the diatom in all years, exclusive of 1980, during which there were slight decreases in phosphate and silica levels. Summer maxima of Fragilaria in each year were accompanied by the exhaustion of silica, the concentration of which was reduced as low as 0.1 mg./l. in Shearwater. The larger summer maxima coincided with more dramatic reduction of silica. Spring peaks of the diatom were also recorded when concentrations of silica were low possibly showing the large utilization of silica by the diatom accompanied by a constant supply from the drainage basin. The maxima and the growth peaks of Fragilaria usually occurred when concentrations of nitrate were already high or rising while those of phosphate were variable.

Practically nothing is known concerning the chemical factors, affecting the growth of F. crotonensis in nature. Active growth of the species in culture was only obtained with concentrations of nitrogen, phosphorus and silica, in excess of those in waters of two lakes in the English Lake District during the periods of active growth of this diatom (CHU, 1942). A marked increase in the numbers of F. crotonensis was synchronous with an increase in the supply of silica from the inflows during summer in Windermere (LUND, 1964). In the present study, the periods of active growth of Fragilaria mostly coincided with already high or increasing concentrations of nitrate, phosphate and silica, indicating the importance of these dissolved nutrients for the growth of the diatom. However,

quite oppositely, the lowest counts of the diatom were also recorded when concentrations of silica were high while those of nitrate and phosphate were high or low at these times. Concerning the active growth and slow multiplication of F. crotonensis, in relation to dissolved nutrients, particularly to silica, the present data would suggest that a factor or a group of factors are also governing the growth of this diatom alongside the dissolved nutrient, studied in this paper.

The autumn increase of Fragilaria was coincident with decreasing temperature and pH level while summer multiplication commenced with increasing temperature and usually with slightly decreasing pH level in this study. However, periods of active growth of the diatom in autumn and summer were synchronous with warm periods within the range 7.9 - 18°C and 12.5 - 18°C respectively. Nevertheless, the actual maximal numbers of the filaments in the autumn periods were achieved within temperaature ranges of 7.9 - 11.5°C and of 16 - 18°C in summer during this investigation.

The highest numbers of the filaments of Fragilaria were recorded in June 1978 and 1980 at very similar temperatures, 16.2°C and 16.4°C respectively. This suggests that the optimum growth of Fragilaria occurred at around 16°C in this study.

APSTEIN (1896) recorded the summer maxima of F. crotonensis at about 20°C in some of the German lakes. WESENBERG-LUND (1904) found the greatest development of the diatom in the lakes of Denmark between 13 and 16°C but it did not flourish above 16°C. F. crotonensis was regarded as having an optimum about 15.5°C in the Illinois river (KOFOID, 1908). RUTTNER (1937b) found

the best growth of F. crotonensis between 7.1 - 14.1°C and FINDENEGG (1943b) between 8 - 14°C but these results were found in the lakes of Austria which being in mountainous regions with cold winters are unlikely to warm to the same extent as Shearwater. There is a distinct possibility that different temperature strains exist.

The present study and the above investigations clearly show that good growth of F. crotonensis is possible within a wide range of temperature but that it could be classified as a warm water species. However, in the present study, a sharp increase in the numbers of filaments of F. crotonensis from 1 to 129 and from 13 to 155 fils./ml. was recorded in 1979 and 1980 when water temperature varied between 3 - 5.5°C. This suggests the possibility of two temperature strains in Shearwater - only experimental studies will reveal the truth or not of this. It is interesting however that in CANTER's work (1982) strains of Fragilaria are found which have a varying susceptibility to fungal attack. ZACHARIAS (1899b) for a similar case, suggested that the increase in available light is the main factor for the great increase in the populations of Asterionella, Fragilaria and Melosira in the Grosser Plöner See in April at a temperature of about 4 - 5°C and whilst this is undoubtedly a most important factor, it may disguise the fact that temperature strains are also present.

The effect of the ice formation in Shearwater on the growth of Fragilaria could not be clearly seen. In 1979, the numbers of filaments were declining before Shearwater was covered by ice and this appeared to be increased by the ice formation. However,

the recovery of the diatom was very quick since the numbers started increasing under the ice and continued to rise until the end of the ice period. It might be noteworthy that there was a decrease in the population after the ice melted and this sequence was again recorded in 1980. It must be pointed out that there was a fungal infection on the diatom population during the ice period in 1980.

The decline in the numbers of Asterionella appeared to be sharper than that for Fragilaria. The autumn growth of Fragilaria usually started before or during the beginning of the vernal increase of Asterionella but whilst Asterionella was still low in numbers, Fragilaria appeared to take advantage of this situation very well and achieved its autumn maxima during such periods of slow multiplication of Asterionella. This may suggest that the rate of cell division of Fragilaria was faster than that of Asterionella in such periods. However as Asterionella accelerated to its vernal maximum during winter, the numbers of Fragilaria were decreasing, and continued to decrease until the end of spring maximum of Asterionella. This suggests that Asterionella may have suppressed the growth of Fragilaria during its vernal increase or that some other factor was slowing the Fragilaria down. Summer development of Fragilaria always coincided with the termination of the spring maximum of Asterionella during this investigation. This once more suggests the growth-dependency of Fragilaria on the competition with Asterionella. This appeared to be one of the main reasons for the onset of the summer maximum of Fragilaria, being late or early in different years during this study.

Similar replacement of Asterionella by Fragilaria was also

observed by GARDINER (1941). In his experimental study, TALLING (1957) was unable to show any inhibitory effects of filtrates of A. formosa and F. crotonensis on one another. LUND (1967) made no definite suggestions why A. formosa is dominant over F. crotonensis in Windermere during winter, although the latter grows as fast as the former from winter to summer in cultures, suspended 1 and 6m.below the surface of Windermere. The same author (1964) attributes the predominance of Fragilaria in relatively small summer crops to its ability to utilize nutrients more efficiently at low concentrations. Phosphate phosphorus and silica are certainly two of these nutrients but the effect of others is unknown.

Summary and conclusions

Two seasonal maxima of F. crotonensis occurred in this study; the summer was larger than the autumn maxima.

F. crotonensis was a persistent component of the phytoplankton in Shearwater since it was present almost throughout these four years of investigation.

Sharp growth peaks and subsequent declines appeared to be characteristic before the actual maxima of the diatom.

The autumn and the summer increase of Fragilaria commenced mostly when the concentrations of nitrate, phosphate and silica were already high or else rising.

The maximum numbers of the diatom in autumn coincided twice with high and once with low concentrations of silica.

However, the actual summer maxima involved the exhaustion of silica in Shearwater. The larger the maximum, the more dramatic the reduction in concentration of silica occurred.

Fragilaria was also absent when concentration of silica was high but nitrate and phosphate could be high or low. This suggests that a factor or group of factors are responsible rather than a single factor.

Rate of active multiplication of Fragilaria was high, maxima being reached within 2 - 3 months.

Fragilaria was tolerant to a wide range of temperature. However, periods of active growth were synchronous with high temperatures (8 - 18°C) but the behaviour of the diatom at similar and different temperatures were quite unpredictable.

Fragilaria appeared to have its optimum growth at around 16°C.

The diatom appeared to be affected by the onset of ice formation in Shearwater since the numbers declined sharply but the recovery of the diatom was very quick and the numbers then increased under ice.

Seasonal periodicity of F. crotonensis appeared to be somewhat dependent on the seasonal cycle of A. formosa, showing the importance of competition between planktonic organisms.

OTHER PENNATE DIATOMS

Some non-planktonic pennate diatoms were also encountered in surface waters of Shearwater. Their occurrence remained low and coincided with the turbulent water.

Amphora ovalis Kütz. occurred in minute numbers mostly coinciding with autumn - spring periods (Fig. 8.). The diatom was more abundant in 1978 and present on most occasions. The numbers did not exceed 7 cells/ml. otherwise numbers remained under 3 cells/ml. The diatom was mostly absent during summer probably due to the stabilisation of the water by thermal stratification.

Occurrence of Cymatopleura solea (Bréb) Smith and a Gomphonema sp. are shown together on the same graph (Fig.8.). These diatoms occurred sporadically in very small numbers, never exceeding 2 cells/ml. They were absent during the large part of this investigation.

Contribution of Gyrosigma sp. to the autumn and spring increase of diatoms was very little indeed (Fig.9.). Maximum of 6 cells/ml. was recorded only once (November). Its appearance in Shearwater was quite sporadic and rare. Appearance of a Pinnularia sp. on two occasions was also shown on the same graph with Gyrosigma sp.

A few small and large species of the genus Nitzschia Hass. appeared in Shearwater after 1978 and their total number is shown together (Fig. 10). The reasons for the absence of Nitzschia spp. before 1978 are obscure. The diatoms were more abundant in 1979 than in the following year. The winter - spring

Fig.8. Upper section: seasonal cycle of

Cymatopleura solea (■) and

Gomphonema sp. (□)

Lower section: seasonal cycle of Amphora ovalis

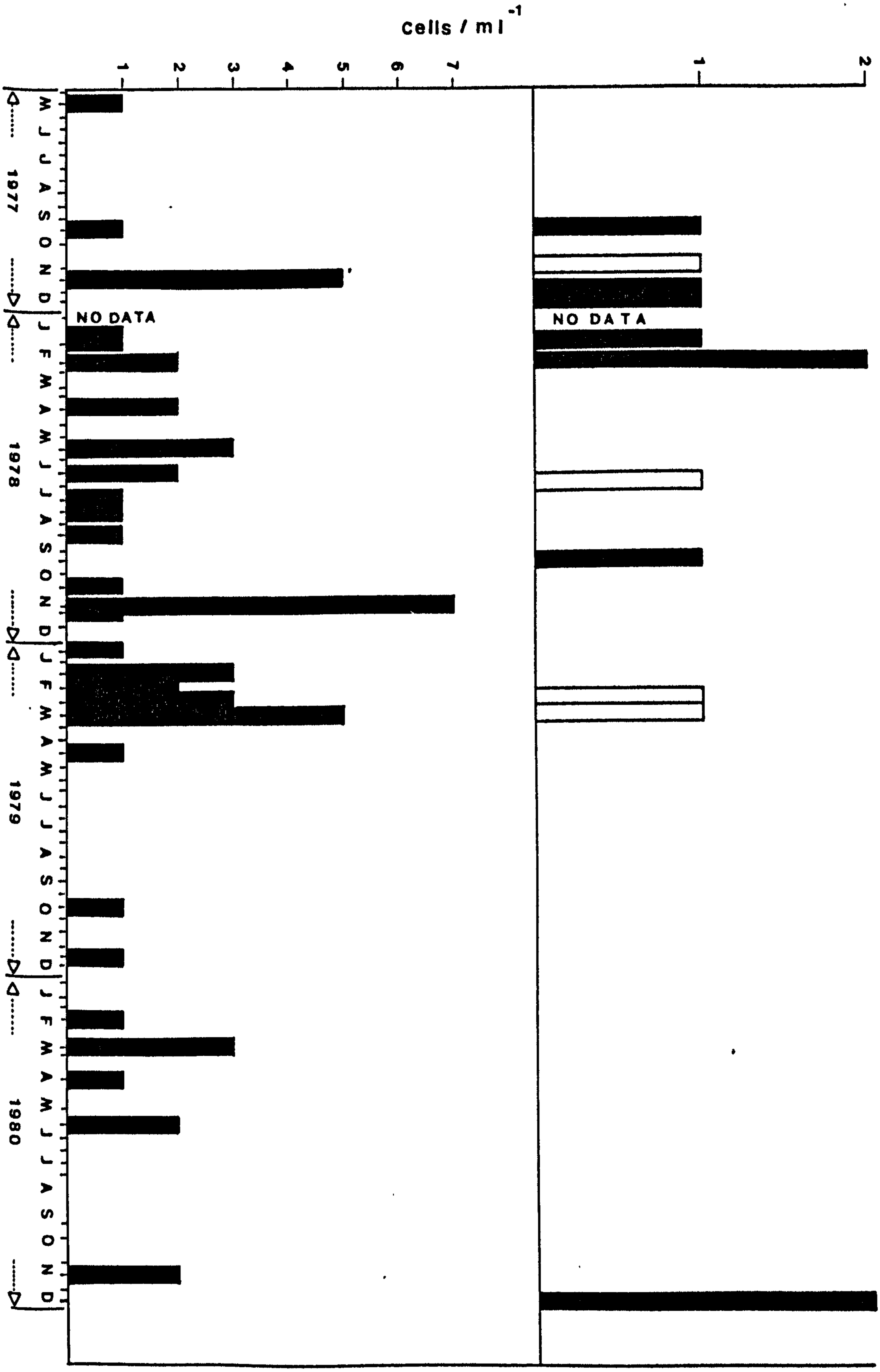

Cells / ml -1
1
2
3
4
5
6
7
1
2
M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980
NO DATA
NO DATA

Fig.9. Seasonal cycle of Gyrosigma sp. (■) and Pinnularia sp. (□).

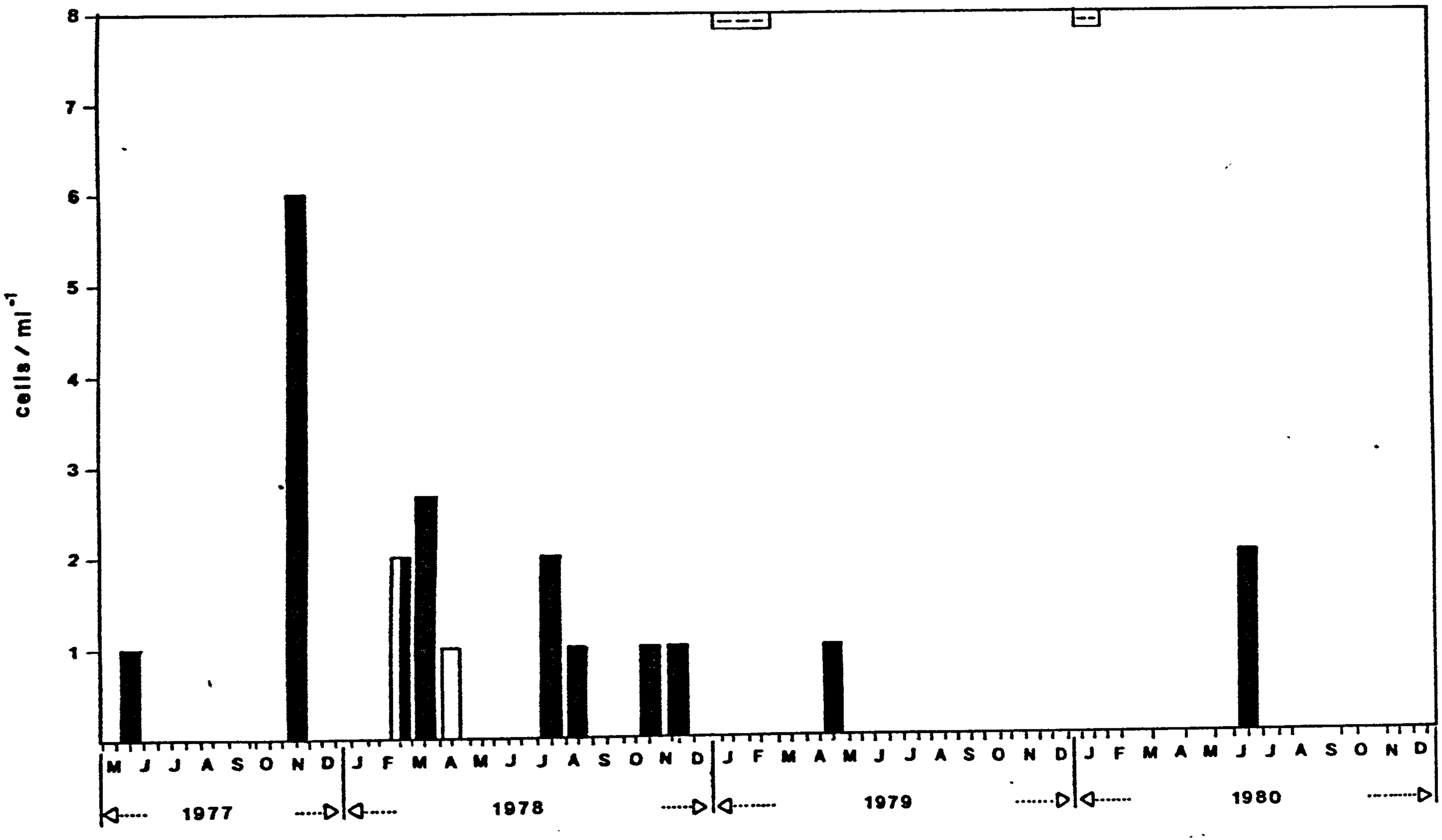
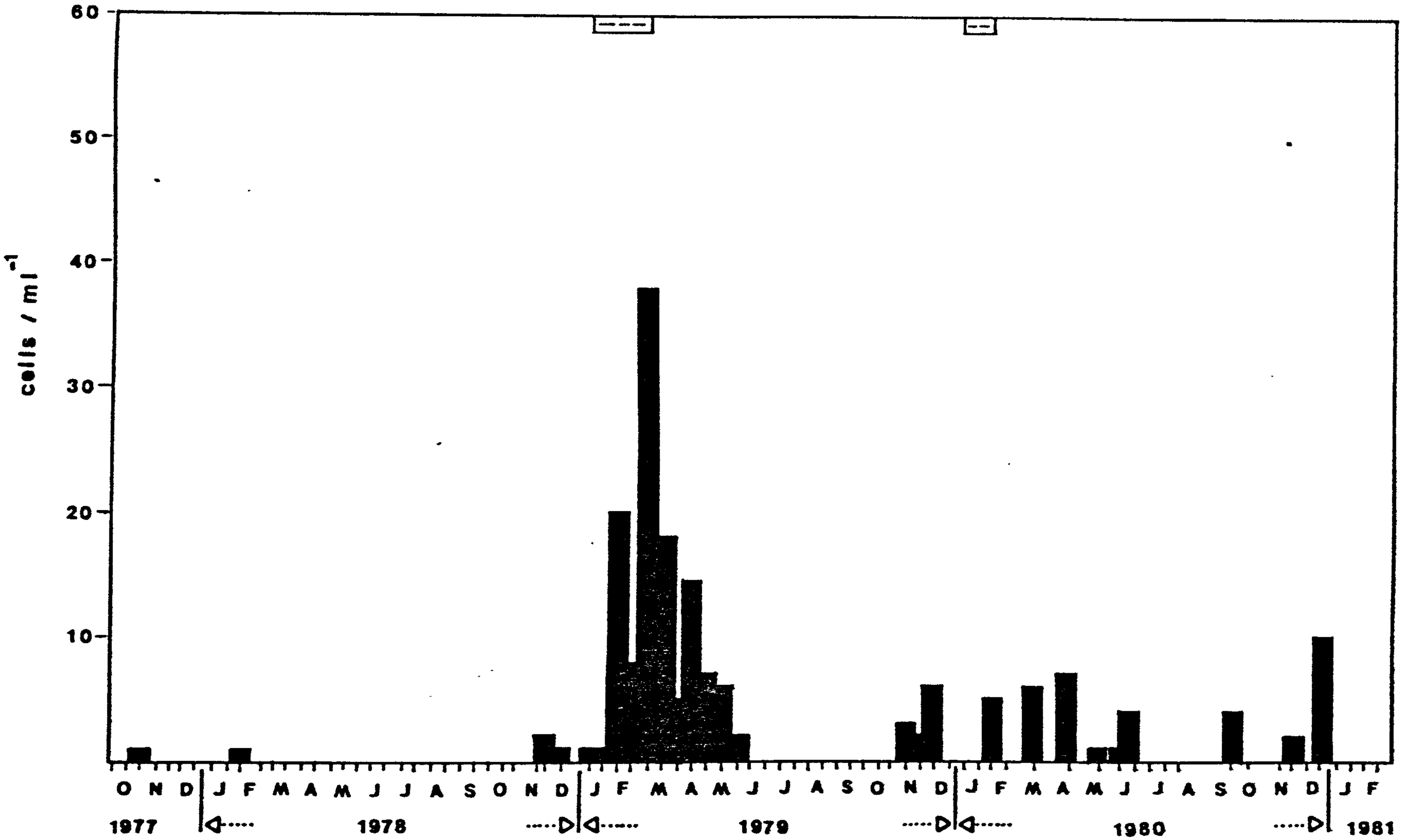

cells / ml⁻¹
8
7
6
5
4
3
2
1
M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

Fig.10. Seasonal periodicity of Nitzschia spp.


60
50
40
30
20
10
cells / ml⁻¹
O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F
1977
1978
1979
1980
1981

(low temperature) period appeared to be more favourable for their growth. A maximum of 38 cells/ml. was recorded only on one occasion coinciding with high silica and nitrate levels. The diatoms were absent during summer.

Synedra ulna (Nitzsch.) Ehr. was also found in the phytoplankton samples of Shearwater (Fig. 11). The numbers were usually low, contributing very little to the autumn - spring development of diatoms in Shearwater. The diatom could be found any time of the year but mostly during autumn. A sharp peak, composed of 77 cells/ml. was the only important feature of its appearance in Shearwater occurring soon after the peak of Nitzschia spp. High concentrations of nitrate and silica probably favoured its growth on that date.

Occurrence of unidentified pennate diatoms in Shearwater is presented in fig. 12. It is apparent that these diatoms were present on most occasions but with usually low numbers. However, they were found more frequently during autumn and spring. The highest number was only 23 cells/ml. recorded in April 1979.

In conclusion, the rare occurrence of these pennate diatoms in Shearwater indicates that the epipelic diatoms are rarely even casual invaders of the eutrophic phytoplankton in Shearwater.

Fig.11. Seasonal periodicity of Synedra ulna.

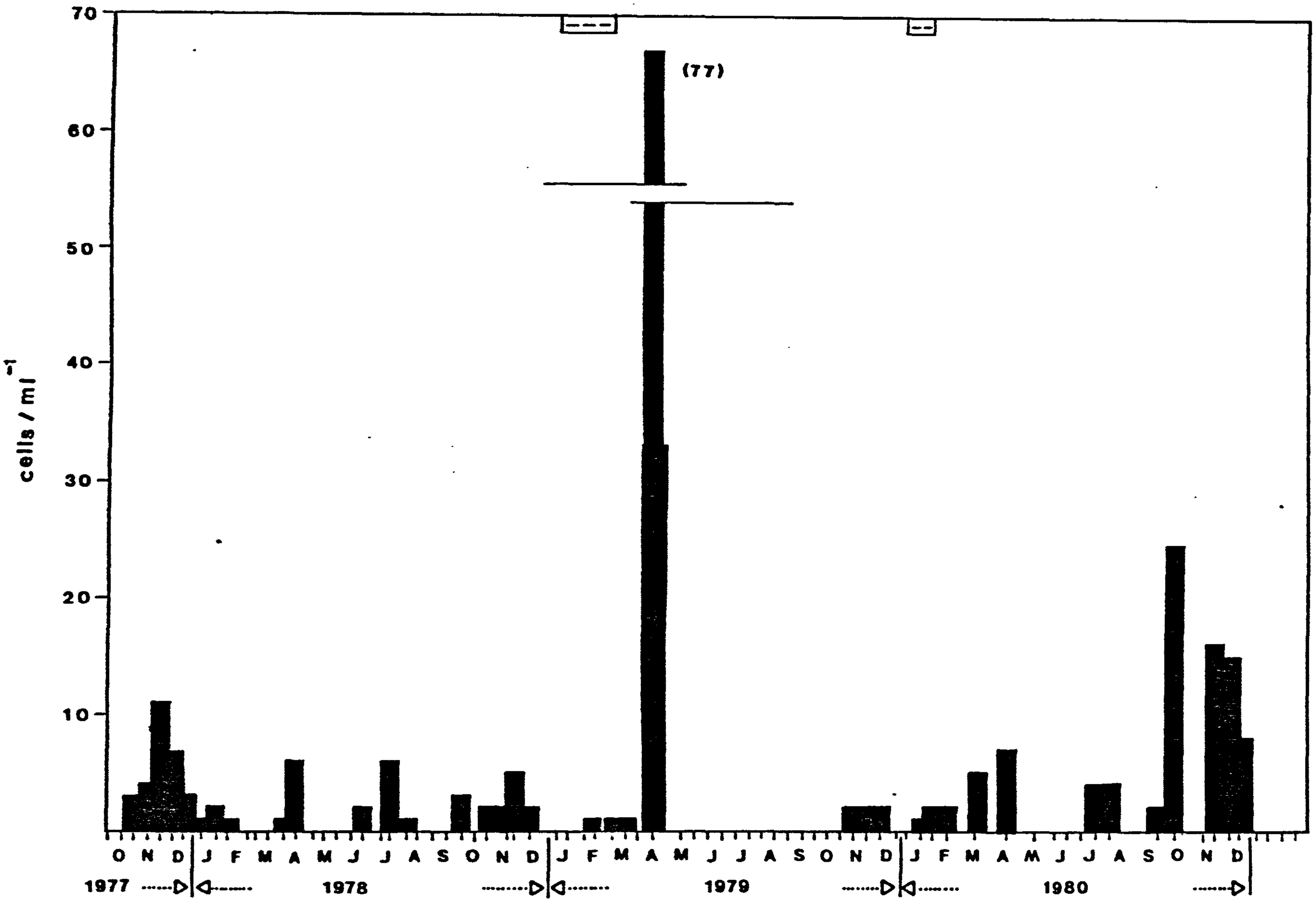
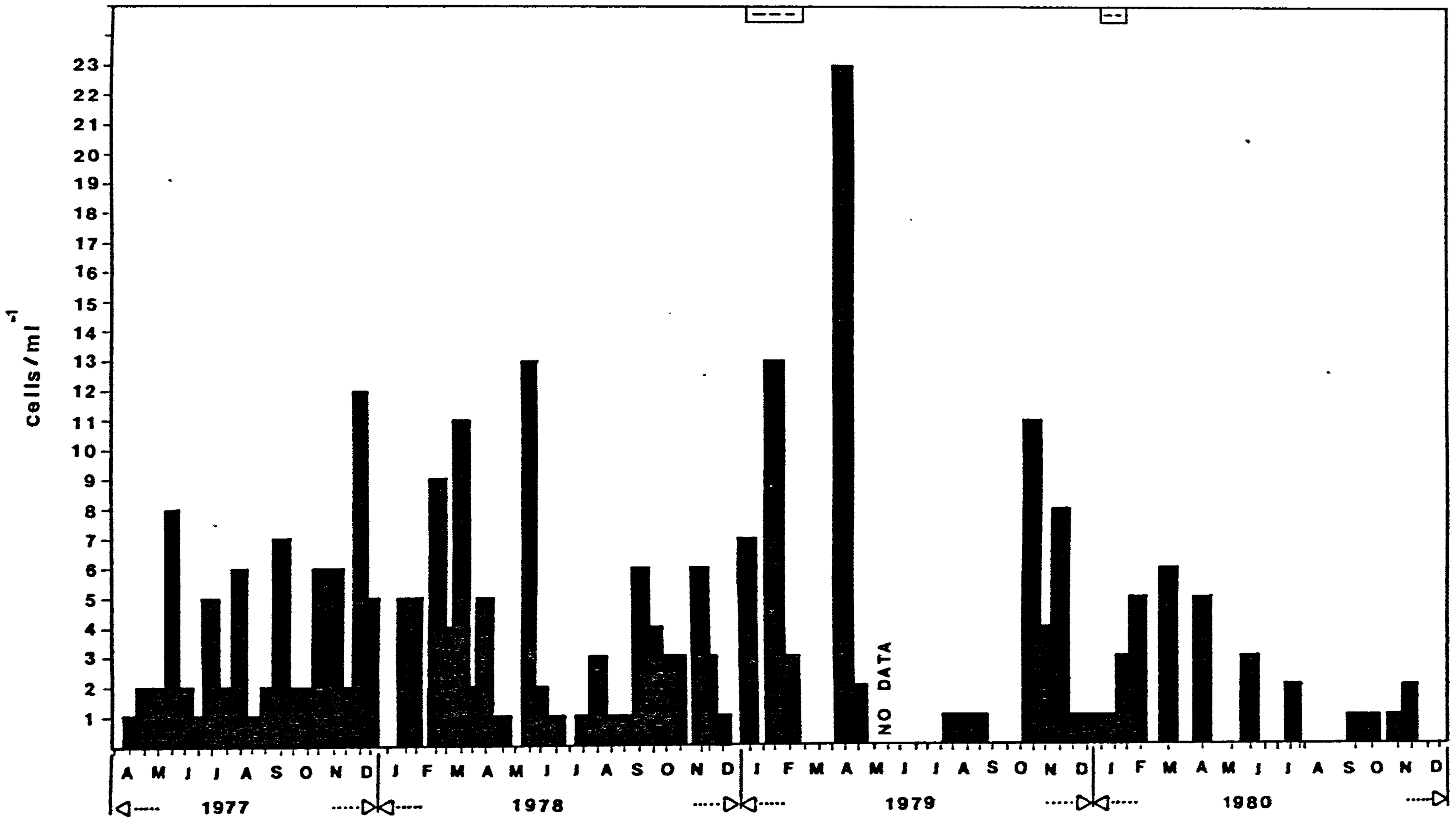

70
60
50
40
30
20
10
cells / ml⁻¹
(77)
O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

Fig.12. Seasonal distribution of unidentified pennate diatoms.


cells/ml⁻¹
23
22
21
20
19
18
17
16
15
14
13
12
11
10
9
8
7
6
5
4
3
2
1
NO DATA
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

CENTRIC DIATOMS

Centric diatoms, particularly Cyclotella and Stephanodiscus were quite conspicuous and responsible for centric blooms while representatives of the genus Melosira occurred in far smaller numbers.

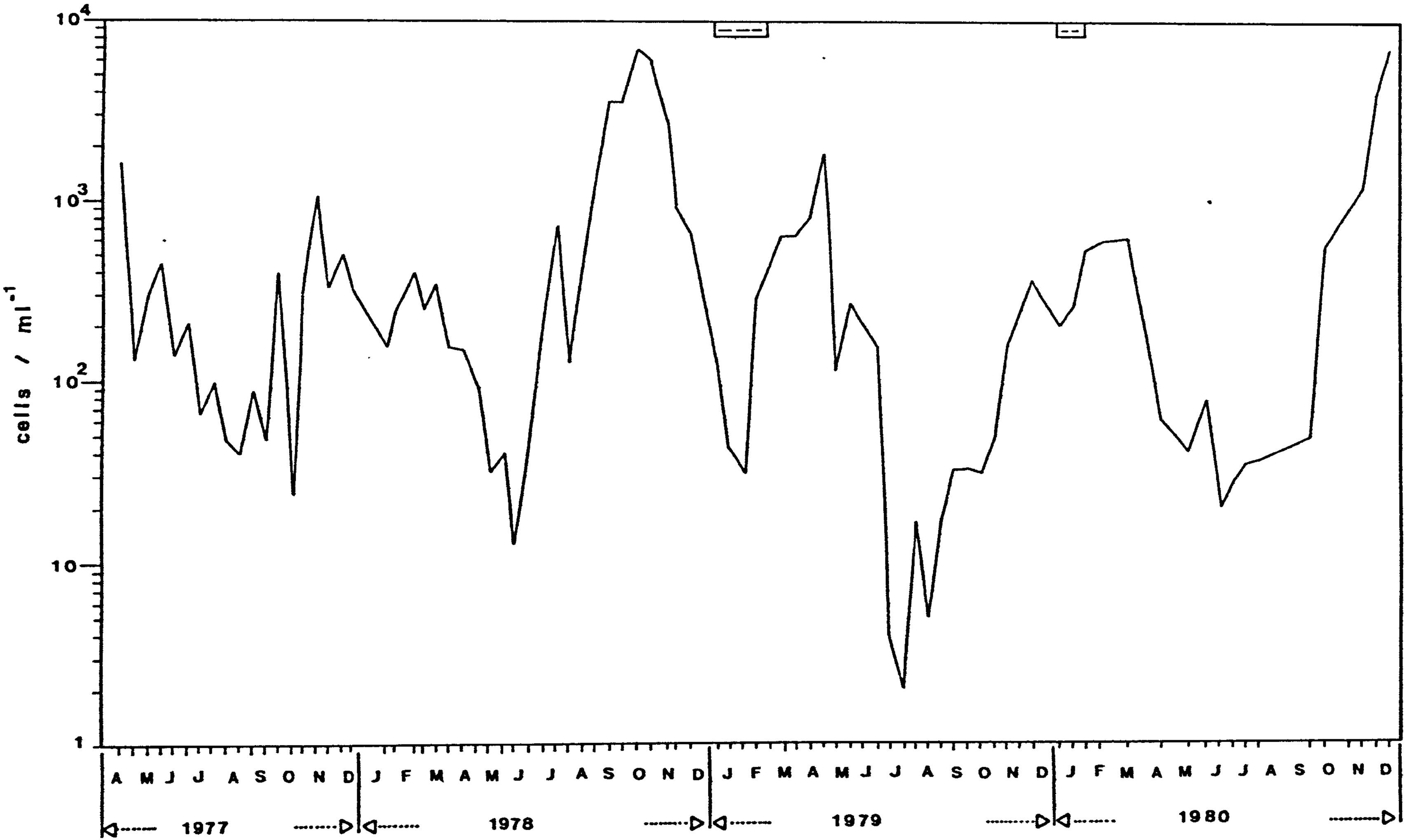
Cyclotella and Stephanodiscus

The genus Cyclotella was represented by two species: C. kützingiana Thwaites and C. meneghiniana Kütz. while Stephanodiscus hantzchii Grun. was the only member of the genus Stephanodiscus in Shearwater. Separate counting of individuals was quite difficult due to their simultaneous occurrence and their similar appearance as live cells. Hence these three species were considered as a single diatom and their total number was plotted on the graph (Fig. 13).

It is apparent from the same figure that these diatoms were present but in different proportions on every occasion during this investigation. They were more abundant in 1978 and 1980 (like F. crotonensis) than in the other two years.

These centric diatoms produced two distinct maxima in Shearwater; one in autumn and another one in spring. In 1977 and 1979 the spring maxima were larger than those of autumn while autumn maxima were 7 - 10 times larger than those of spring in the other two years. However, the spring maximum in 1978 and the autumn one in 1979 were not as conspicuous as in the other years. These diatoms were generally in small numbers from late

Fig.13. Seasonal variations in the total numbers of Cyclotella and Stephanodiscus.


cells / ml-1
10^4
10^3
10^2
10
1
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

spring to early summer but brief growth peaks also occurred during such periods.

Commencement of the autumn development of the diatoms varied in each year, starting in October in 1977, August 1978/79 and September in 1980. Maximum autumnal numbers in 1977 and 1978 were achieved earlier (October/November) than in the two following years (August) following 1 to 4 months of active growth. The size of the autumn maxima also varied from year to year and a smaller maximum in one year was always followed by a much larger one in the next year regularly during this investigation. Maxima in 1979 and 1980 were 7 and 18 times as large as the ones recorded in the other two years.

The spring increase of the centric diatoms commenced in winter (December - January) and maxima occurred within 1 - 3 months of active multiplication. Spring maxima were reached in February 1978, April 1979 and March 1980. Although there is no complete data for 1977, maxima appeared to be in April. Size of the spring maxima also showed the same characteristic features as the autumn maxima, i.e. smaller maxima were always followed by larger ones. Spring maxima in 1977 and 1979 were far larger in size than those in the other two years. In addition an immense number of minute unidentified centric diatoms were observed in April 1979 and January - February 1980 which are not shown in the graphs of Cyclotella and Stephanodiscus. Numbers of these minute centrics were approximately 20,000 - 40,000/ml. indicating the great speed of multiplication when the conditions are favourable.

These centric diatoms always declined steadily after the autumn and spring maxima were achieved. Duration and degree of declinations differed in each year.

Late spring-summer periods were usually associated with much lower numbers of centric diatoms compared to those recorded in the other periods of the year. However, a few small peaks also occurred during this less numerous period of the diatoms. In addition, a sharp growth peak in July 1978 indicated that centric diatoms could also increase fast during such periods if the conditions favour their growth. However, the lowest numbers were recorded in October 1979 and June - July in the following years.

Seasonal periodicity of Cyclotella and Stephanodiscus in Shearwater resembled or differed in part or fully from early ecological studies of these genera in other lakes. COLDITZ (1914) recorded a maximum of Cyclotella meneghiniana in September which was then replaced by C. hyalina which appeared at the end of October, producing maxima in November. BIRGE & JUDAY (1922) found that Cyclotella sp. contributed to spring maxima of Stephanodiscus in many years. Species of Cyclotella were reported to be dominant or of quantitative importance in Lake Constance, Switzerland, producing maxima in August. CHANDLER (1940, 1942a, b, 1944) found Cyclotella and Stephanodiscus tended to be autumnal while RILEY (1940) and HUTCHINSON (1944) reported Stephanodiscus and great numbers of minute round cells, probably partly Cyclotella, taking part in the spring maximum of diatoms. FINDENEGG (1943b) reported the maximum of Cyclotella during June - July. KARIM (1965) found

that S. hantzchii was almost invariably present and often prominent in Abbot's pool showing a seasonal distribution similar to Shearwater. Development of Stephanodiscus hantzchii in autumn and spring in Crose Mere was observed by REYNOLDS (1973) as in this study.

Growth of Cyclotella and Stephanodiscus in Shearwater was synchronous with high as well as very low temperatures indicating that the development of these diatoms was not influenced by temperature . Maxima occurred within a range of 5.2 - 17°C and the size of the maxima did not show a relationship with the temperature.

Ice formation did not appear to affect the growth of centric diatoms in Shearwater. A maximum was reached under ice in 1980 supporting the view of SWALE (1964) that Stephanodiscus hantzchii can build up large numbers in slow-flowing water. However, the autumn decline of centric diatoms continued under ice in 1979, and a sharp increase in the numbers coincided with the ice melt.

COLDITZ (1914) recorded the maxima of Cyclotella meneghiniana in autumn at a temperature between 9.3 - 11.3°C which is a narrower range than that recorded in this study. Autumnal growth of Cyclotella and Stephanodiscus over a temperature of 10 - 23°C was reported by CHANDLER (1940, 1942, a, b, 1944). SWALE (1964) also observed the maximum development of Stephanodiscus hantzchii in spring and autumn; findings in harmony with Shearwater. However the present data suggests that the seasonal fluctuations of Cyclotella and Stephanodiscus may be independent of seasonal climate conditions since high numbers

were found during the greater part of the year. A relatively slow sinking rate of Stephanodiscus hantzchii was observed in culture by SWALE (1964) and the persistant presence of Cyclotella and Stephanodiscus may be attributed to this in this study as well.

Chemical data for Shearwater is shown in fig. 3 and correlations may be made with the seasonal cycle of centric diatoms (Fig.13).

Autumn increase of Cyclotella and Stephanodiscus showed a clear relationship with nitrate and silicate. Concentrations of both dissolved nutrients were generally increasing while the influence of phosphate was complicated since sharp increases and equal declinations occurred during such periods.

Increasing concentrations of nitrate also coincided with the spring development of the diatoms while concentrations of phosphate and silica increased or declined sharply.

Although maxima of Cyclotella and Stephanodiscus were achieved at high as well as low levels of silica, a fall in concentration always occurred when the maxima were reached. Nitrate was still at high levels during maximal numbers of the diatoms, however, declines also coincided during some maxima. Phosphate either decreased or rose when maxima were achieved.

During the autumn and spring declines of the diatoms, silica returned to the water whilst nitrate was mostly decreasing and phosphate declined or increased.

In conclusion nitrate and silica supported the growth of Cyclotella and Stephanodiscus with increasing or high concen-

trations while phosphate did not appear to be a limiting factor.

It is of interest that the development of Asterionella formosa and Cyclotella/Stephanodiscus coincided more or less with the same periods. These diatoms did not appear to affect growth of one another since periods of increase were simultaneous.

Summary and conclusions

Cyclotella and Stephanodiscus were persistent members of the phytoplankton in Shearwater, being present throughout the year. This shows the high degree of tolerance by these diatoms to the environmental changes.

Two maxima were produced by Cyclotella and Stephanodiscus in Shearwater and a smaller maximum in one year was followed by a larger one in the next year in the case of both autumn and spring maxima.

Autumn development of the diatom appeared to be faster than that of spring. Two large maxima occurred in autumn.

Numbers of the diatom were lower during late spring - summer periods. However it was also observed that they can grow very well during such periods.

Growth of Cyclotella and Stephanodiscus did not appear to be influenced by the temperature.

Development of the diatoms usually coincided with increasing or high levels of nitrate and silicate while influence of phosphate was confusing.

The seasonal cycles of Cyclotella and Stephanodiscus were more or less similar in Shearwater.

The genus Melosira C. Agardh.

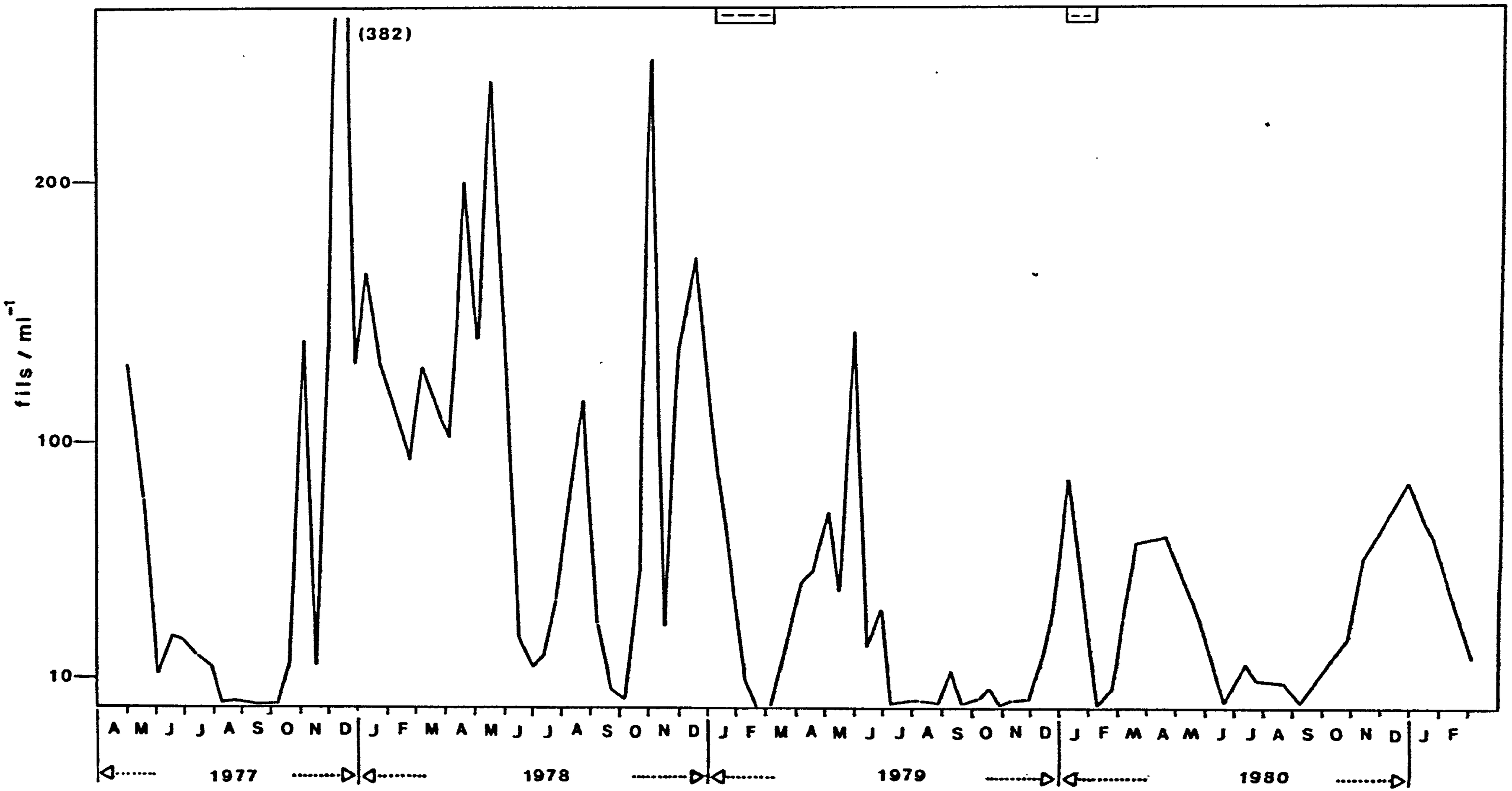
The genus Melosira was the only genus represented by four species in Shearwater. These were M. ambigua (Grunow) Müller, M. granulata (Ehr.) Ralfs, M. granulata var. angustissima Müller and M. varians C.A. Agardh.

Seasonal fluctuations in the number of filaments of M. ambigua (Fig. 14) and M. varians (Fig. 16) are shown on separate graphs while a joint graph represents M. granulata and M. granulata var. angustissima (Fig. 15). It is apparent from these graphs that M. ambigua and M. granulata were persistent diatoms in the phytoplankton of Shearwater, occurring on most occasions while the other two Melosira spp. appeared on fewer occasions. M. ambigua was overall the most abundant of all.

M. ambigua produced two annual growth maxima coinciding with the autumn and spring development of previously mentioned planktonic diatoms. The number of filaments of M. ambigua appeared to be higher in autumn than in spring showing the enhanced growth of the diatom in the autumn. During winter the number of filaments was low whilst in summer the diatom was virtually absent. However, a sharp growth peak was also recorded once during the summer period (1978) so that the possibility of growth is affirmed. The highest autumnal number of filaments were recorded during October - November while those of spring were achieved in April. A maximum of 380 fils/ml. in October 1977 was exceptional and was the highest number recorded.

M. granulata was present more frequently than M. granulata var. angustissima, but both occurred in more or less the same numbers. However autumn 1980 was exceptional in that an extreme

Fig.14. Seasonal periodicity of Melosira ambigua.


(382)
200
100
10
fills / ml^{-1}
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F
1977
1978
1979
1980

growth peak of M. granulata occurred (68 fils./ml.) while the variety was represented by only 5 fils./ml. Generally both diatoms occurred in low numbers remaining under 15 fils./ml. throughout this investigation exclusive of the peak in 1980. However, small peaks of both diatoms could occur at any time of the year without any regular pattern.

M. varians was the least conspicuous member of the genus and the filaments were encountered only during typical autumn and spring developments of diatoms in Shearwater. The autumnal numbers of M. varians were higher than those of the vernal although the diatom was rare even during such periods. During summer, the diatom was totally absent probably due to formation of thermal stratification. Maxima of 7 and 8 fils./ml. was recorded only on two occasions, otherwise the number of filaments generally remained under 3 fils./ml.throughout this investigation.

RICE (1938b) found M. granulata var. angustissima to be prominent in autumn while FLINT (1949/50) and LUND (1962a) observed M. granulata and M. granulata var. angustissima to be abundant in the summer. FINDENEGG (1943b) reported that M. granulata usually occurred at the time of the autumnal maxima but that it could also produce a small secondary maximum in May, whilst REYNOLDS (1973) reported regular large populations of the diatom in summer. However in this study M. granulata and M. granulata var. angustissima occurred in low numbers,only the former producing a maximum coinciding with the autumn - winter period. M. ambigua and M. agassizii were found by LUND (1954) to be most abundant under isothermal conditions whereas the number of M. ambigua in Shearwater was either low or virtually absent

Fig.15. Seasonal cycle of Melosira granulata (———)
and M. granulata var. angustissima (- - - - - - -)

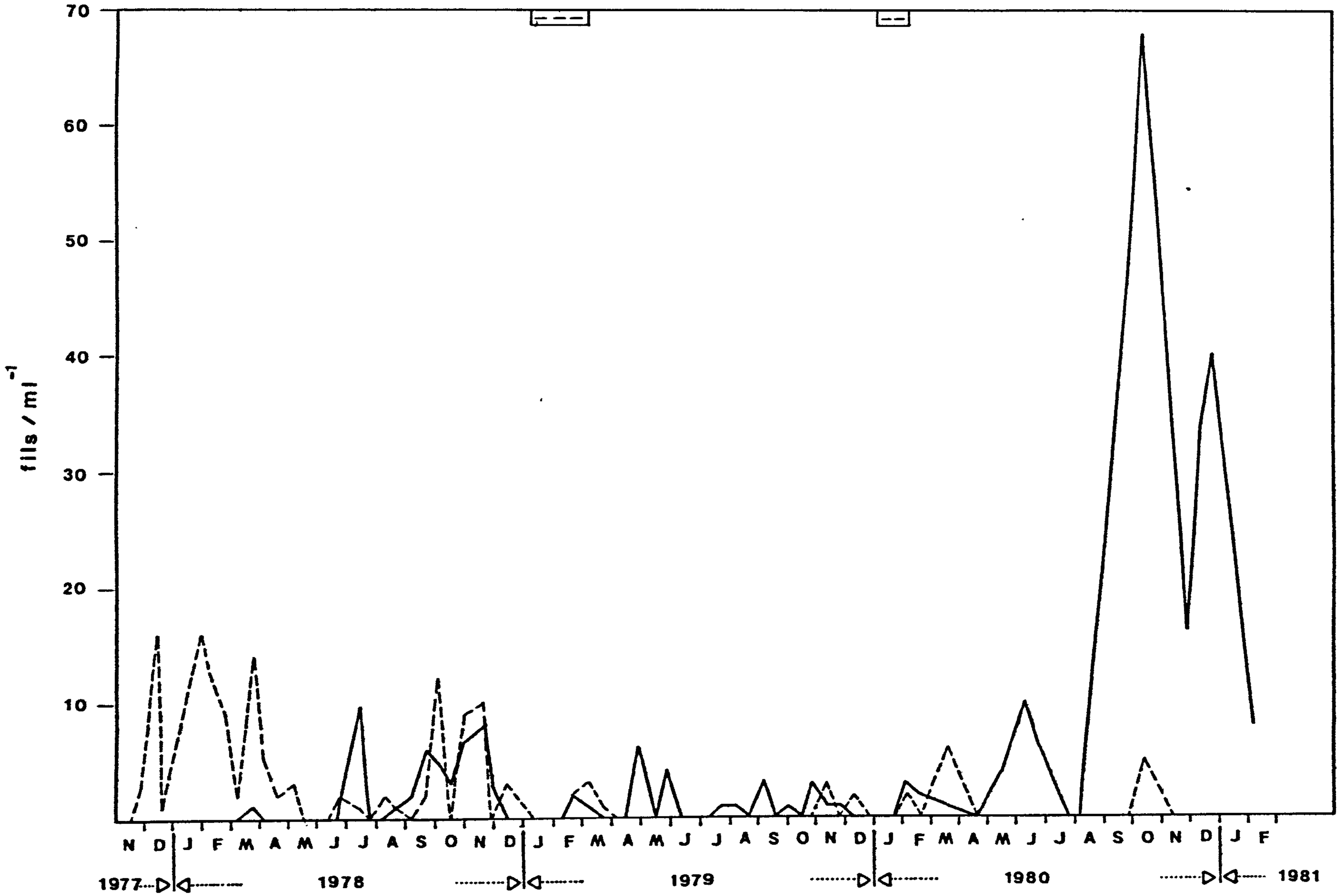
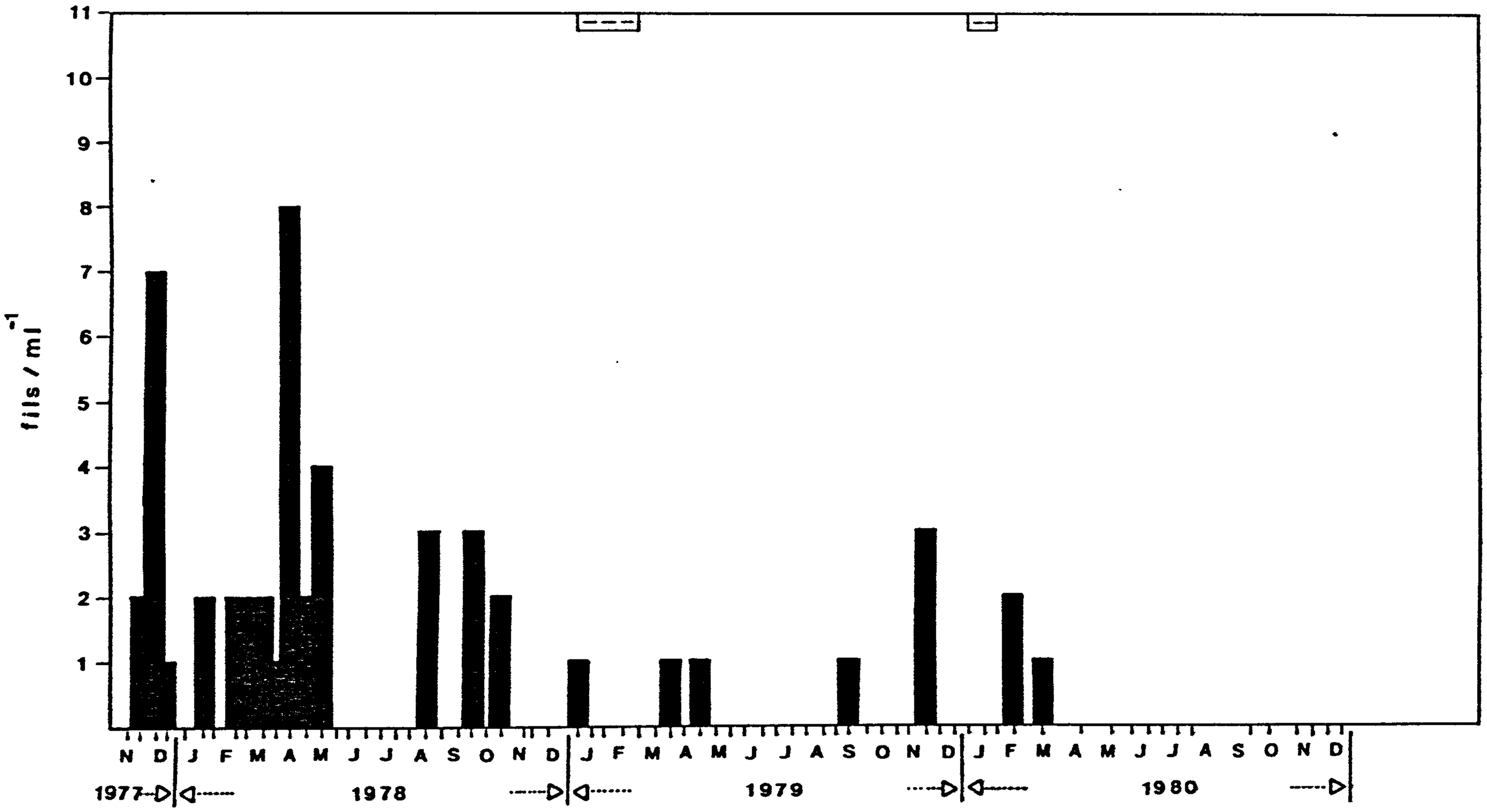

70
60
50
40
30
20
10
fils / ml^{-1}
N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F
1977
1978
1979
1980
1981

Fig.16. Seasonal cycle of Melosira varians.


11
10
9
8
7
6
5
4
3
2
1
fils / ml^{-1}
N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

during such periods.

Melosira spp. showed a high degree of tolerance to changes in temperature during this study. Increasing numbers were synchronous with decreasing as well as increasing temperatures. However, very cold and very high temperatures did not appear to favour their growth. Ice formation in Shearwater coincided with low or virtual absence of Melosira spp. apart from M. granulata var. angustissima which actually increased under the ice in one year but was absent during the second ice period in Shearwater. This suggests that the growth of Melosira spp. in Shearwater is affected by the ice formation. Re-suspension of filaments occurred after the ice melted in Shearwater. WESENBERG-LUND (1904, 1908) recognized that the high autumnal population of Melosira disappeared under the ice and he attributed this to the fact that the filaments tend to sink in the relatively non-turbulent water and they are suspended when the ice breaks, a suggestion which probably holds true for Shearwater. WEST & WEST (1912b) recorded M. granulata producing maxima at temperatures between 1.7 and 4.9°C and WEST (1909) recorded maxima between 18.5 and 23.3°C. Whilst only maximum of M. granulata was produced in Shearwater, they occurred at temperatures between 6.8 and 20°C.

Development of M. ambigua, M. granulata and M. granulata var. angustissima mostly coincided with increasing or high levels of dissolved silica. However on a few occasions silica was reduced to very low levels. During the active growth of these diatoms, concentrations of nitrate and phosphate were also usually increasing. Small growth peaks of M. varians were synchronous with high levels of nitrate and silicate while phosphate was decreasing or rising.

RICE (1938b) found the growth of M. granulata var. angustissima was favoured by high concentrations of nitrate while KARIM (1965) reported its good growth at low as well as high silica levels. Both observations are in harmony with the present data.

Summary and conclusions

The genus Melosira was represented by four species in Shearwater and M. ambigua was overall the most abundant.

Melosira spp. were most abundant during typical autumn and spring increase of the diatoms. Two maxima occurred regularly for M. ambigua; the autumnal one was larger than that of spring. Other species were sparse and did not produce conspicuous maximum. However, M. granulata produced a maximum once in autumn.

Melosira spp. were quite tolerant to changes in temperature. However, their growths were affected by ice formation since they were generally absent during ice periods.

Development of Melosira spp. was favoured by increasing or high levels of dissolved nitrate, phosphate and silica.

CYANOPHYCEAE

Blue-green algae were major components of the summer - autumn phytoplankton in Shearwater and usually reached their maximum numbers in the periods of declining or low numbers of diatoms and green algae.

Aphanizomenon flos-aquae (Lyngb.) De Brébisson.

A. flos-aquae was the most conspicuous member of the blue-green algae in Shearwater and usually produced large blooms during summer andrarely during autumn.

It is apparent from the seasonal cycle of A. flos-aquae (Fig.17) that the occurrence of the alga was limited to summer - autumn months. The alga was more abundant in 1977 and 1979 and the number of filaments was very similar in these years.

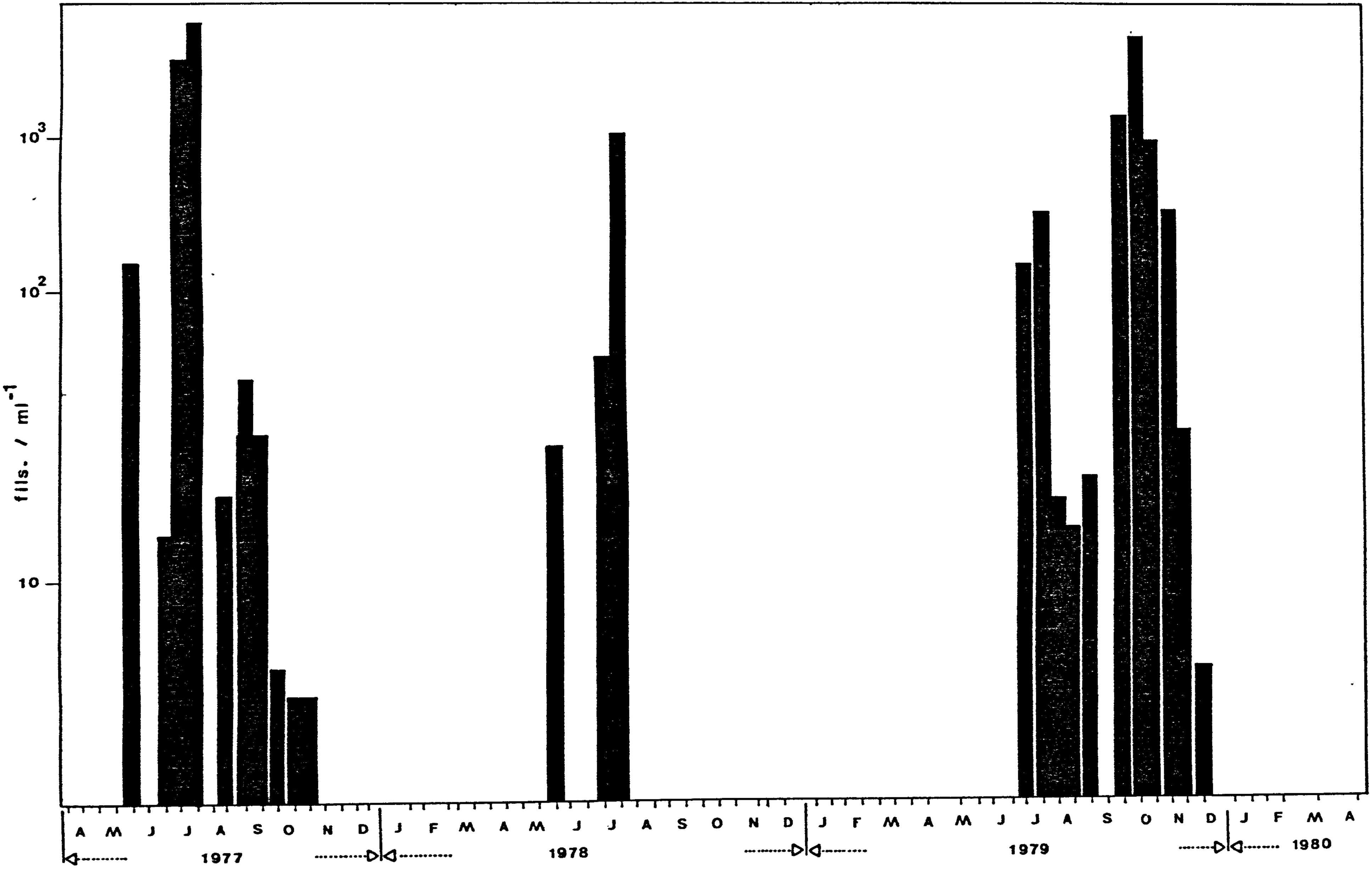
In 1977, the alga appeared in the samples by late spring and reached its summer maximum of 3404 fils./ml.in June. Autumn development of the alga was less,producing small maximum of 81 fils./ml.in September.

The alga occurred on fewer occasions and with less numbers in 1978. There was only a summer maximum (1050 fils./ml.) occurring again in July.

In 1979, the summer maximum (463 fils./ml.) was much smaller than the autumn maximum (2992 fils./ml.) but its duration was the longest. No data for the growth of A. flos-aquae was obtained for 1980.

NAUWERCK (1963) also found A. flos-aquae occurred in July whilst REYNOLDS (1971) recorded spring and summer development

Fig.17. Seasonal periodicity of Aphanizomenon flos-aquae.


10^3
10^2
10
fils. / ml^{-1}
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A
1977
1978
1979
1980

of the alga in the Shropshire Meres. The latter author also observed that A. flos-aquae increased very slowly at temperatures below 10°C but a more rapid increase occurred between 10 and 15°C. The periods of active growth of A. flos-aquae in Shearwater were recorded at temperatures between 7 and 20°C and the best growth between 10 and 20°C. This suggests that high temperatures favour the development of A. flos-aquae.

Observation of REYNOLDS (1971) that the main periods of increase of Aphanizomenon and Anabaena populations were accompanied by large reduction in nitrate level was partly supported by the present data. Increasing populations of A. flos-aquae were synchronous with decreasing but also rising concentrations of dissolved nitrate and phosphate.

Summary and conclusions

Aphanizomenon flos-aquae was a typical summer - autumn form in the phytoplankton, producing large maxima.

High temperatures (10 - 20°C) favoured the growth of the alga while the influence of dissolved nitrate and phosphate remained obscure since periods of main increase were coincident with high as well as low concentrations.

Anabaena Spiroides Klebahn.

A. spiroides occurred only during late spring - summer periods and was less numerous than A. flos-aquae.

The numbers of A. spiroides in 1977 and 1979 were far greater than in the other two years (Fig. 18). Two important maxima were produced by the alga; a maximum of 189 fils./ml. in May 1977 and a second one of 710 fils./ml. in August 1979. Small peaks in July - August also occurred in the other two years. Development of A. spiroides was either slightly earlier than that of Chroococcalean blue-green algae or they were simultaneous.

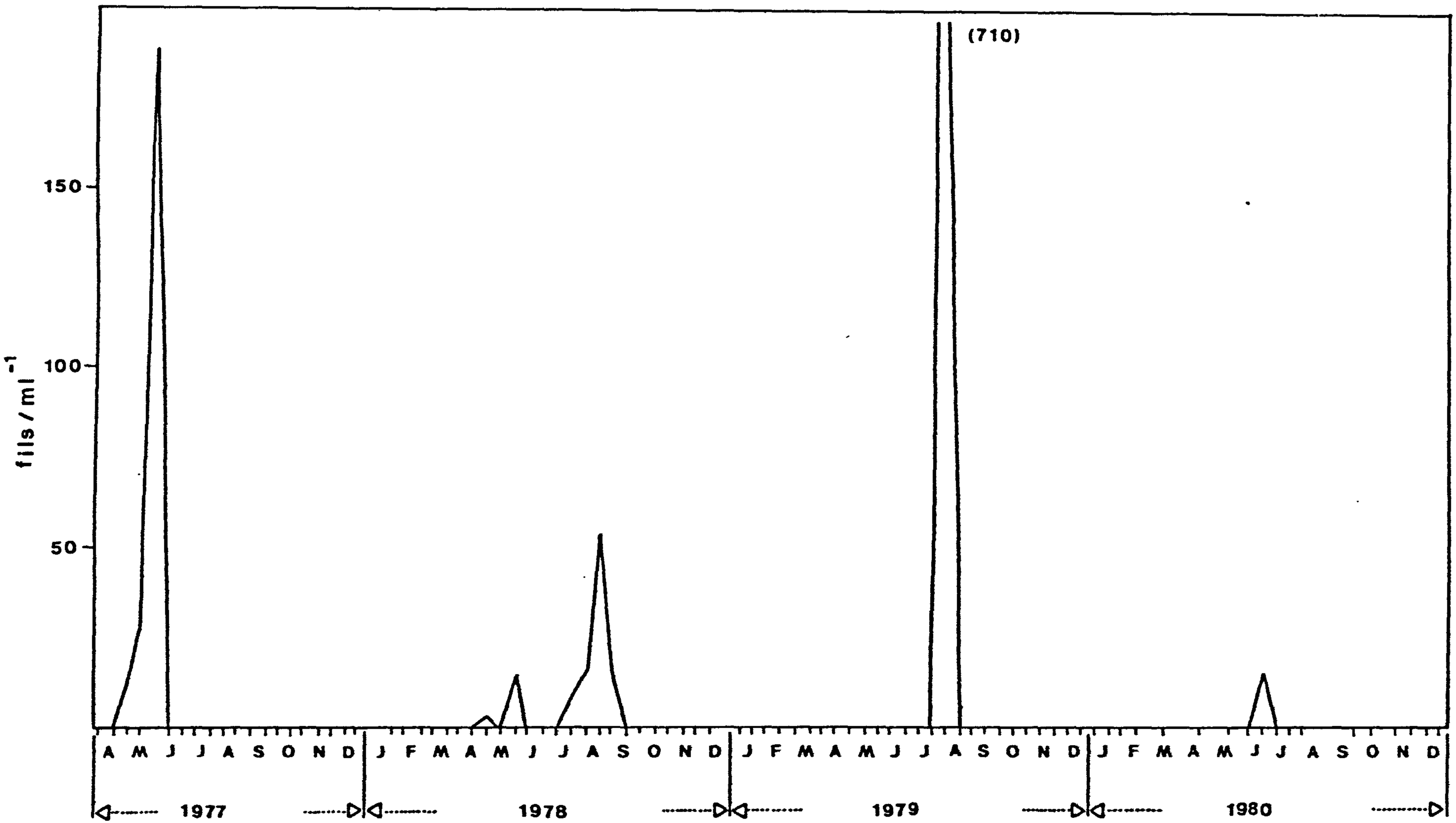
The present data is, partly, in agreement with REYNOLDS (1971) who also found Anabaena tends to be most abundant in late spring. The periods of increasing A. spiroides population were coincident with high temperatures (16 - 18°C). The active growth of A. spiroides coincided with low or decreasing concentrations of nitrate and rising or high levels of phosphate, thus supporting the view of REYNOLDS (1971).

Summary and conclusions

A. spiroides increased during late spring - summer periods and produced two important maxima.

The alga occurred at temperatures of 16 to 18°C.

Fig.18. Seasonal distribution of Anabaena spiroides.


(710)
150
100
50
fils / ml⁻¹
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

CHROOCOCCALES

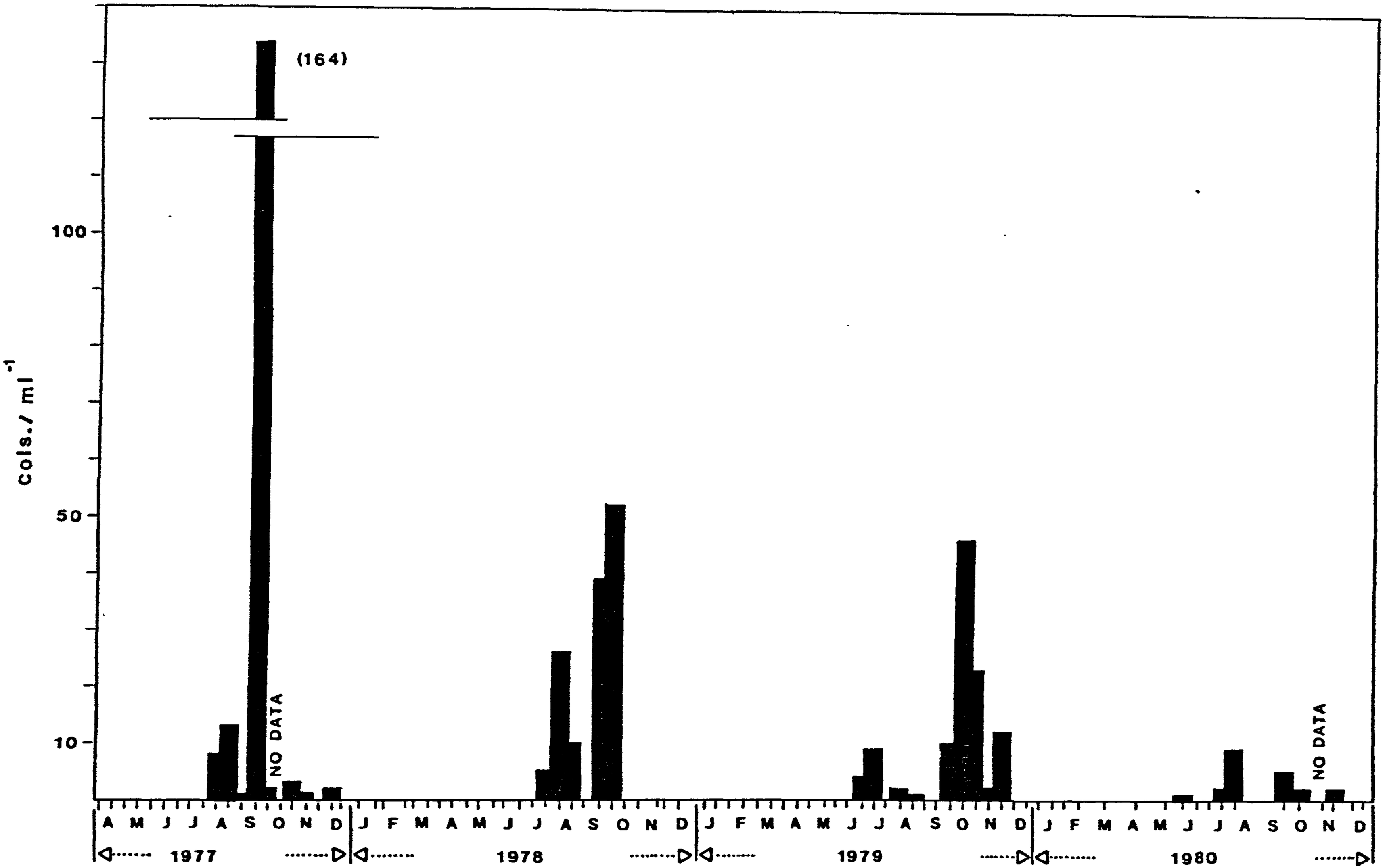
Coelosphaerium naegelianum Unger, Gomphosphaeria naegelina (Unger.) Lemm. and Microcystis aeruginosa Kuetz; emend. Elenkin were the major members of summer - autumn plankton in Shearwater and their occurrence more or less coincided each year. The colonies of M. aeruginosa were found more frequently than those of the others, and were usually co-dominant in Shearwater with A. flos-aquae. They changed the colour of surface water to a greenish colour and gave rise to thick oily layers on the edges of the lake.

The total number of colonies for Chroococcalean blue-green algae is shown in Fig. 19. It is apparent that their major growth was in the autumn months. In fact, summer development of the alge was unimportant during this study. The autumn maximum of 164 cols./ml. in 1977 was exceptionally large clearly indicating the dominant role of these algae especially considering the number of cells within the sizeable colonies. In the two following years, there was a great drop in the number of colonies and in 1980 they failed to develop into appreciable numbers.

The present study differed from that of REYNOLDS (1971) who found M. aeruginosa and C. naegeliana to be abundant during summer at temperatures between 16 - 21^{o}C, slightly higher than the temperature at which they were abundant in Shearwater.

GERLOFF et al. (1952) considered that in many lakes nitrogen was more likely to be a limiting factor for the growth of Microcystis than was phosphorus. REYNOLDS (1971) also suggested nutrient limitation for M. aeruginosa due to the fact

Fig.19. Seasonal distribution of total numbers of *Coelosphaerium naegelianum*, *Gomphosphaeria naegeliana* and *Microcystis aeruginosa*..


(164)
100
50
10
cols./ ml-1
NO DATA
NO DATA
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

that the alga failed to develop at low levels of nitrate and phosphate. However, the development of Chroococcalean blue-green algae in this study was generally favoured by increasing or high levels of nitrate and phosphate.

Summary and conclusions

C. naegelianum, G. naegeliana and M. aeruginosa occurred more or less in the same periods and their occurrence in autumn was more conspicuous than in summer.

Their growth was favoured by high temperatures and increasing levels of nitrate and phosphate.

CHLOROPHYCEAE

Green algae particularly members of the Chlorococcales were important components of the phytoplankton in Shearwater. In general they were present throughout this investigation in different proportions, however,reaching their maximum only during summer. Some species were present throughout the year while others gave rise to distinct phases at certain times of the year.

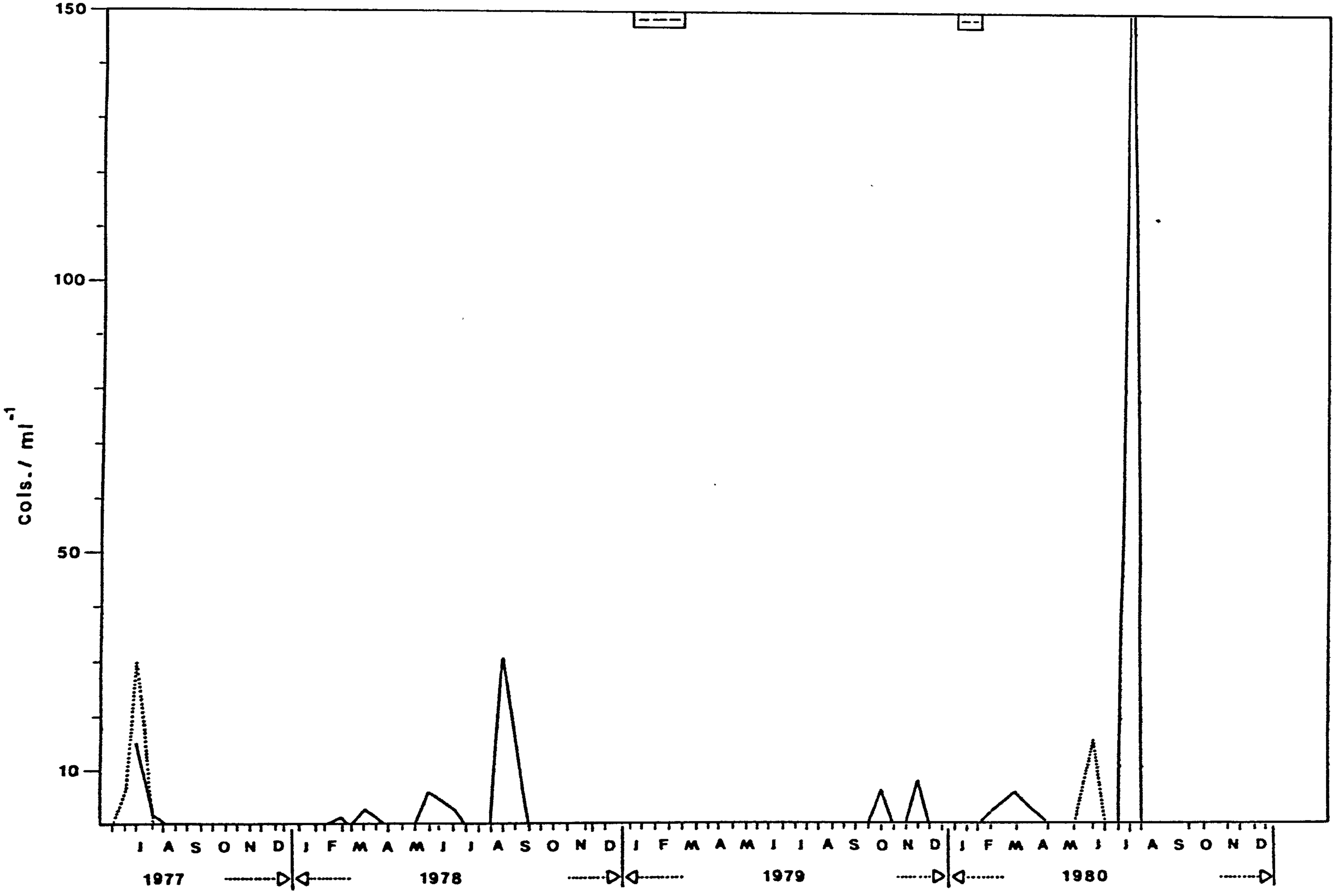
Volvocales

The presence of Pandorina morum (Müll.) Bory, Volvox aureus Ehr. and V. globator Linn. was detected almost from the beginning of this investigation; Volvox was only rarely recorded.

Small numbers of colonies of Pandorina morum were encountered during late winter - spring and the autumn period but occurrence was sporadic (Fig. 20). However, the alga reached its highest number for short times during summer (e.g. 152 cols./ml. in July 1980) indicating that the periods of high illumination and temperature favoured growth.

A summer increase of Pandorina morum is well known in many lakes. FRITSCH & RICH (1913b) observed that low temperatures in the summer favour the growth of P. morum and RICE (1938b) suggested that the alga was favourably affected by sunshine and low temperatures. However, in the present study the alga showed its best growth at high temperatures in the summer and periods of low temperatures were coincident with small numbers. The

Fig.20. Seasonal periodicity of Pandorina morum (———) and Volvox spp. (.....)


150
100
50
10
cols./ ml-1
J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

present study is in harmony with that of HODGETTS (1922) who found P. morum starting development during spring and becoming plentiful during summer. Spring maximum of the alga were recorded by RAO (1955) who relates the abundance of the alga to high illumination and low rainfall.

The appearance of Volvox aureus and V. globator was quite irregular since they were recorded only on a couple of occasions(Fig. 20). It is apparent from the same figure that Volvox spp. were typical summer forms in Shearwater.

It is obvious that development of Volvocacean algae in Shearwater was not controlled only by climatic conditions since their occurrence and numbers varied very much under more or less similar climatic conditions. Effect of dissolved nutrient on their growth was also complicated, records being during low or high concentration phases.

In conclusion, the present study would suggest that there are clearly other factors, not revealed by the present study, which at least play a part in controlling the occurrence of these algae.

Summary and conclusions

Volvocean algae, Pandorina morum, Volvox aureus and V. globator were sporadic typical summer forms in Shearwater.

CHLOROCOCCALES

The members of this order were the most conspicuous of all the Chlorophycean algae and were also major components, particularly during summer.

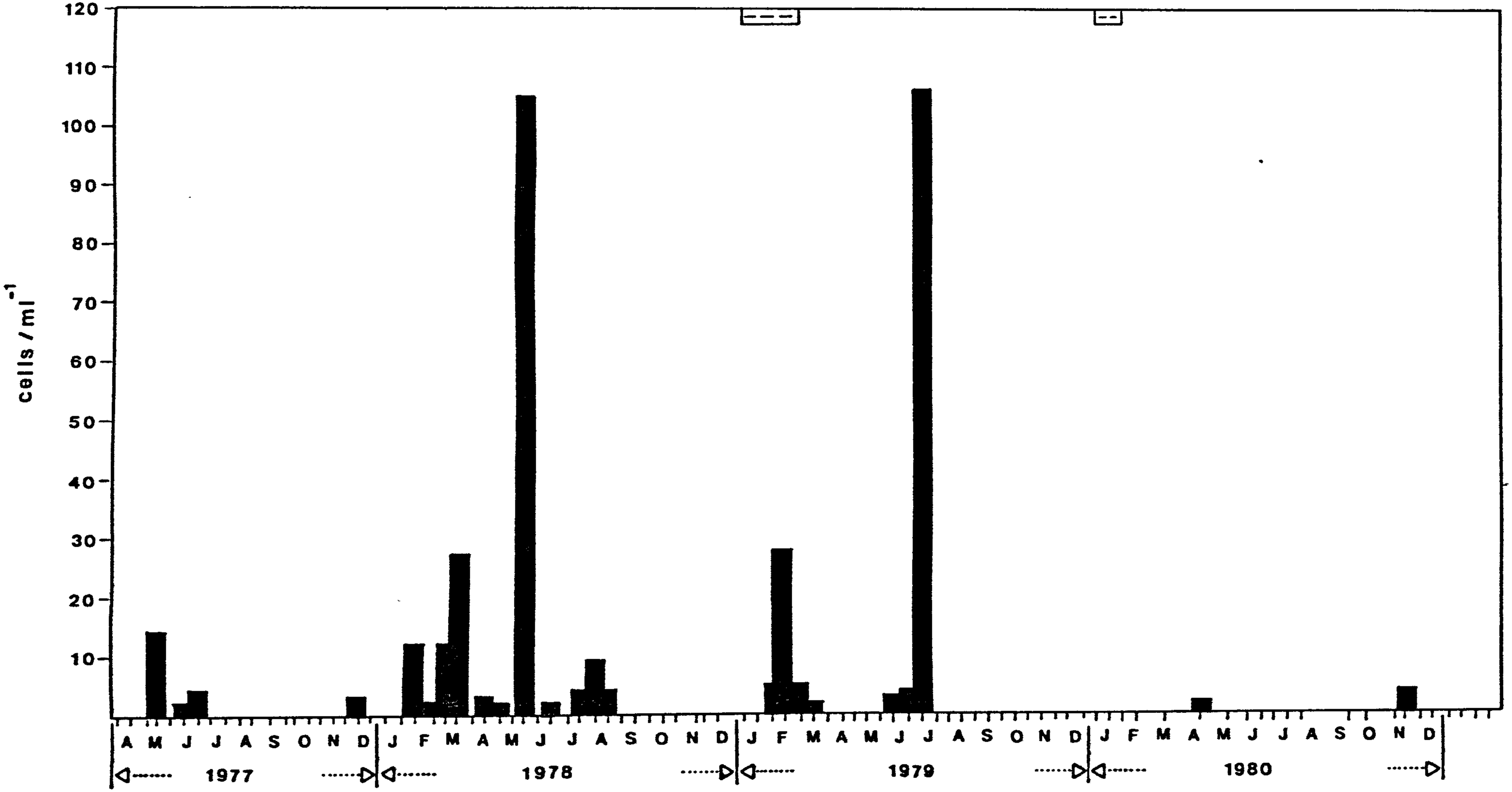
Ankistrodesmus falcatus (Corda) Ralfs

The occurrence of A. falcatus was negligible in 1977 and 1980 while regular occurrence and more abundant numbers of the alga were recorded in the other two years. (Fig. 21)

Two development phases of A. falcatus were observed in the more numerous years of 1978 and 1979. The alga was found with small numbers in winter and soon after produced its small maximum either in late winter or early spring. The second development of A. falcatus commenced by late spring and reached its higher maxima either in the same period or mid-summer. Two successive summer maxima of the alga were composed of more or less the same numbers. A. falcatus was then sporadic, absent for the remainder of the year.

HODGETTS (1921) found A. falcatus to be adapted to moderately high temperatures (11 - 14°C) and RAO (1955) found Ankistrodesmus to be prominent in the spring. Spring development of A. falcatus was also recorded in the present study and the alga was also increasing under the ice. This suggests that A. falcatus can adapt to very cold temperatures as well. RODHE (1948) suggested that A. falcatus requires high phosphate and nitrate but here the development and maxima of the alga coincided with high as well as low concentrations of nitrate

Fig.21. Seasonal periodicity of Ankistrodesmus sp.


120
110
100
90
80
70
60
50
40
30
20
10
cells / ml^{-1}
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

and phosphate. In addition, similar physico-chemical conditions also occurred in 1980 but the alga was virtually absent. This suggests that the growth of A. falcatus is also controlled by some other undetermined factor.

Summary and conclusions

Two development phases of Ankistrodesmus falcatus occur; the alga is more numerous in late spring - summer than in late winter - early spring.

Some undetermined factors appeared to control the growth of the alga alongside nitrate and phosphate concentration.

The genus Coelastrum Naegeli

The genus Coelastrum was represented by two species and only C. reticulatum (Dang.) Senn produced large maxima while the occurrence of C. microporum Naegeli was relatively unimportant (Fig. 22).

C. microporum was absent in Shearwater during the larger part of this investigation and only small numbers of coenobia being encountered during autumn and summer. C. reticulatum was present on more occasions and also occurred in small numbers during autumn and summer and produced two small maxima (June 1978 and May 1980).

These maxima and a small one in 1979 were achieved at temperatures between 16 - 18°C.

Temperatures between 20 - 25°C were regarded as limiting thermal optima for Coelastrum by RODHE (1948).

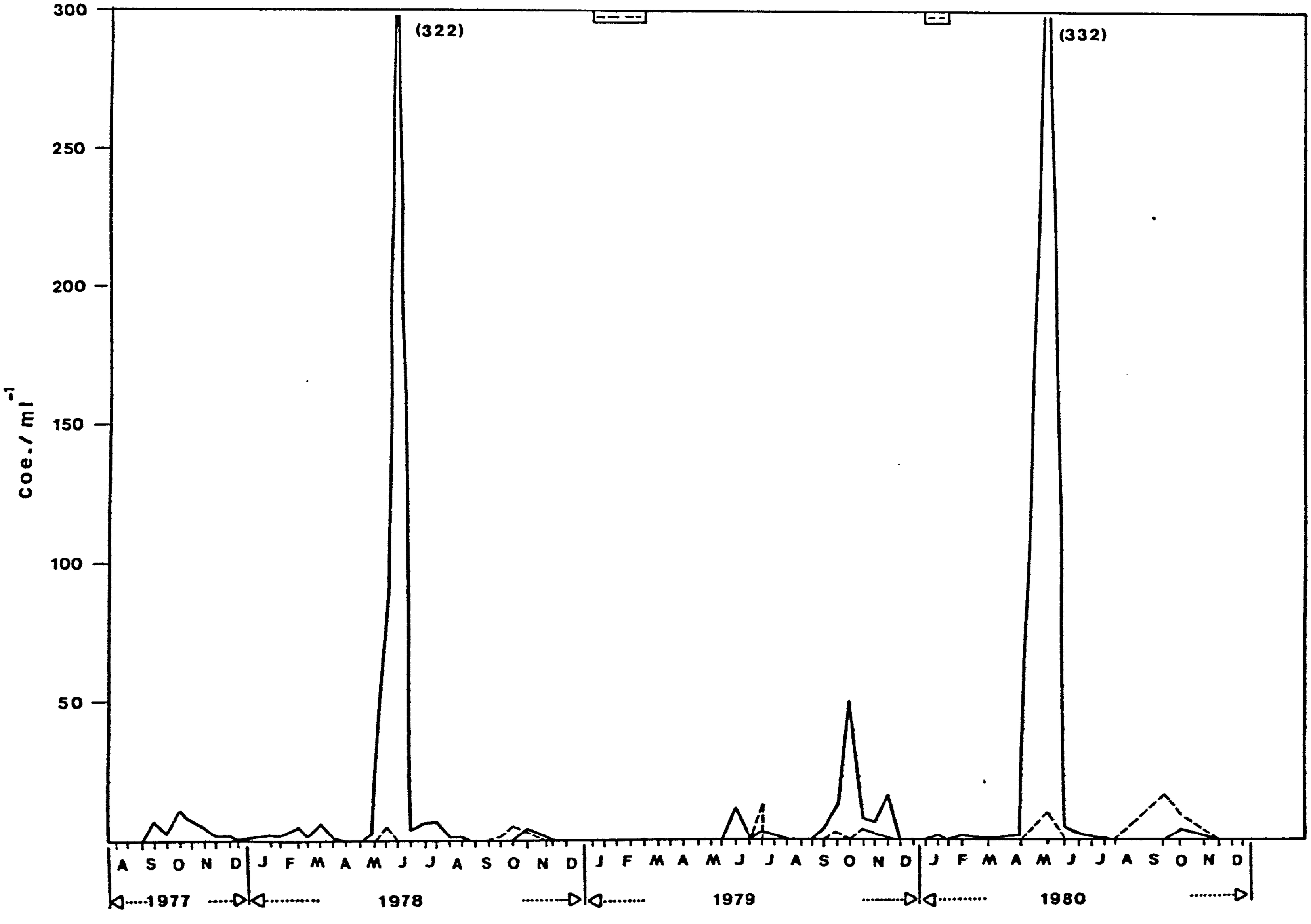
Correlation between occurrence of Coelastrum spp. and dissolved nutrients was also complicated in this study but the two maxima coincided with decreasing concentrations of dissolved nutrients.

Summary and conclusions

Apart from two growth maxima of C. reticulatum the growth of C. microporum and C. reticulatum was unimportant in Shearwater.

High temperatures appeared to favour the growth of these green algae.

Fig.22. Seasonal periodicity of Coelastrum reticulatum (————) and C. microporum (--------).


300
250
200
150
100
50
Coe./ ml^{-1}
(322)
(332)
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

Dictyosphaerium pulchellum Wood

Seasonal distribution of the number of colonies of Dictyosphaerium pulchellum is shown in Fig. 23 . It is clear from the same figure that the numbers were small and the alga preferred spring - early summer months and that its numbers in 1978 were relatively more significant than in other years. However, a few colonies were also found irregularly during autumn. Two growth peaks (46 cols./ml.in June 1978 and 26 cols./ml. in April 1980) were the only important feature of D. pulchellum during this investigation.

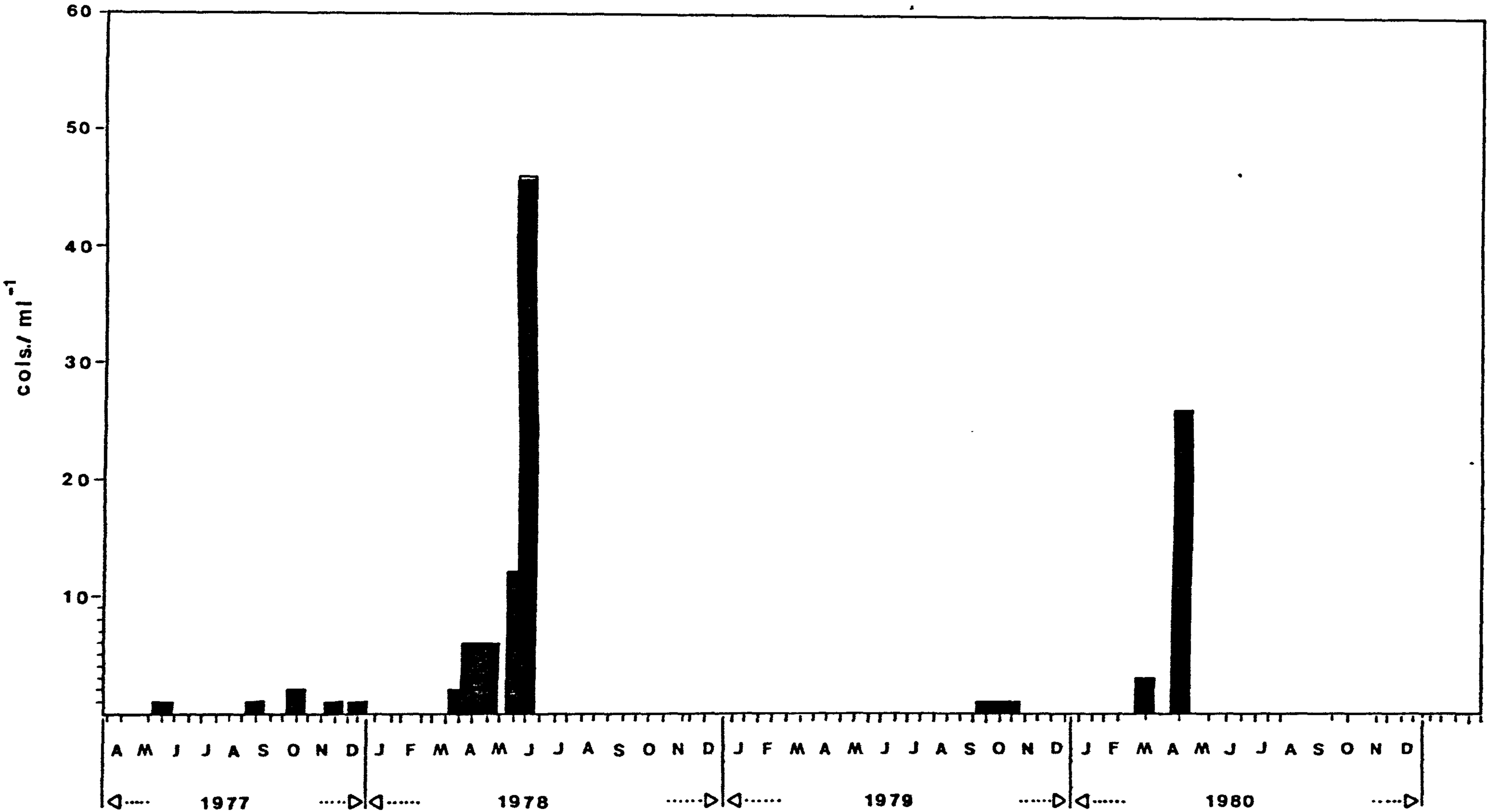
The seasonal cycle of D. pulchellum in Shearwater differed from that recorded by KARIM (1964) in Abbot's Pool who found the alga growing better during autumn. The present data is also in disagreement with THOMASSON (1963) who reported that Dictyosphaerium forms a considerable part of the plankton in the cold and rainy season. Best growth of the alga occurred at temperatures of 12 and 18^{o}C in this study whilst nutrients were high but decreasing.

Summary and conclusions

Dictyosphaerium pulchellum was absent on most occasions and occurred sporadically during this investigation.

The alga favoured spring - early summer months and its occurrence coincided with high temperatures and high concentration of dissolved nutrients.

Fig.23. Seasonal distribution of Dictyosphaerium pullchellum


60
50
40
30
20
10
cols./ ml^{-1}
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

Oocystis lacustris Chodat

The occurrence of O. lacustris was restricted to mid spring - summer months in Shearwater (Fig.24). However, sporadic occurrence of the alga with small numbers during autumn was also observed but it was completely absent during winter. O.lacustris was more abundant in 1978 and 1980 than in the other two years. Although Oocystis has been recognised as an oligotrophic type (HUTCHINSON, 1967) it must be considered as eutrophic from this study.

The growth pattern of O. lacustris was more or less the same in three years and the alga remained in the phytoplankton samples for 3 to 5 months. Two conspicuous maxima (43 cells/ml. in June 1978 and 76 cells/ml. in May 1979) were recorded throughout this investigation although small peaks occurred more frequently during its presence in Shearwater.

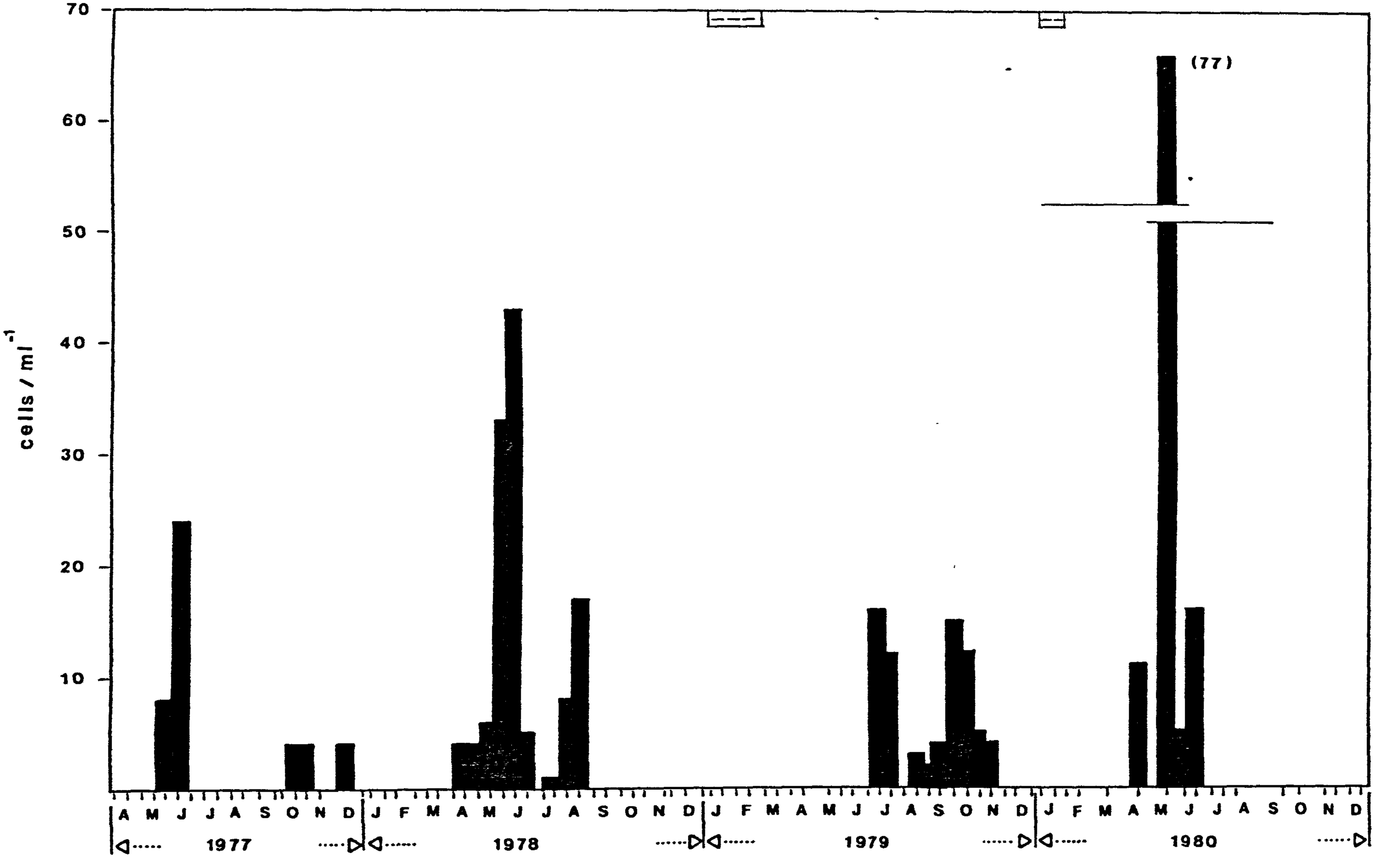
The distribution of cell numbers in the colonies during its occurrence is shown in table

	1 celled	2 celled	4 celled	8 celled
Number of col.counted	93	3	42	9

Table:2. Total number of cells in colonies of Oocystis lacustris

It is clear from the above table that 1 celled and 4 celled colonies were present more frequently than 2 and 8 celled colonies in Shearwater. The highest number of cells per colony was coincident with the largest maximum of O. lacustris.

Fig.24. Seasonal periodicity of Oocystis lacustris.


70
60
50
40
30
20
10
cells / ml^{-1}
(77)
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

Growth of O. lacustris was synchronous with temperatures between 10 - 18°C and the two maxima occurred when temperatures were 16°C and 18°C. Periods of growth of O. lacustris did not correlate with nitrate and phosphate since concentration of these dissolved nutrients were either increasing or declining.

It was a striking coincidence that the peaks of Coelastrum reticulatum, Dictyosphaerium pulchellum and Oocystis lacustris were simultaneous in 1978. This suggests that the growth of these green algae was favoured by the same factors.

Summary and conclusions

The growth pattern of Oocystis lacustris was almost the same in three successive years and its occurrence was limited to mid-spring/summer months.

The alga reached to optimum growth at temperatures of 16 - 18°C.

Concentrations of nitrate and phosphate were decreasing when the maxima of the alga were achieved.

The genus Pediastrum Meyen

The genus Pediastrum Meyen was represented mainly by P. boryanum (Turp.) Menegh. and to a much lesser extent by P. duplex Meyen.

P. boryanum was one of the most conspicuous members of the phytoplankton in Shearwater and it occurred in appreciable numbers throughout this investigation, while P. duplex occurred only for a short time with much lesser numbers (Fig.25).

The seasonal cycle of P. boryanum in Shearwater showed that the development of the alga was not limited to a season since it was almost invariably present in the phytoplankton. However, total cell numbers were higher in autumn and summer periods.

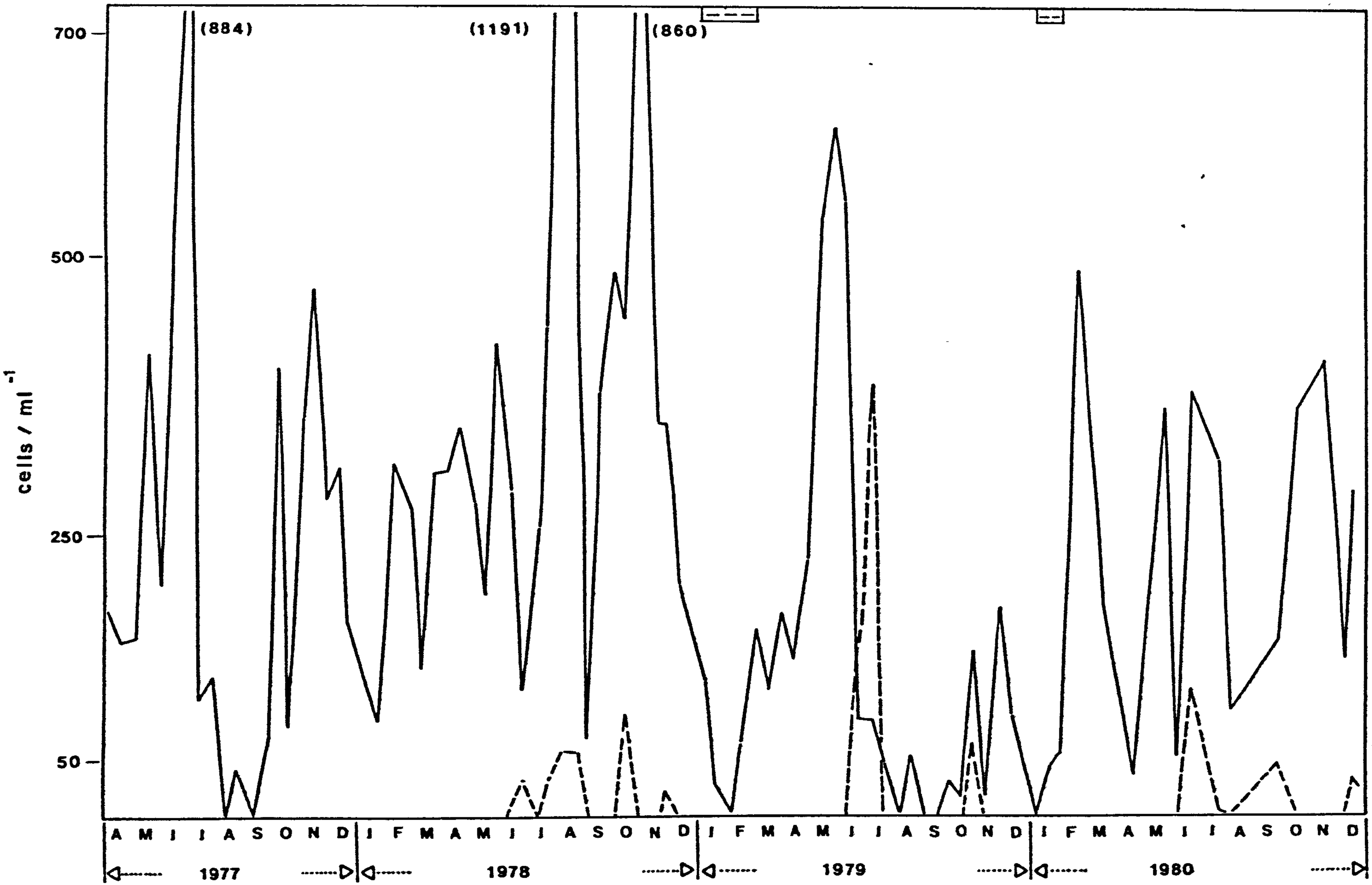
In 1977, P. boryanum produced two maxima and the summer maximum (June) was twice as large as the autumnal one (November). The alga was virtually absent during August- September.

In 1978 two large maxima also occurred and summer maximum (August) was once more by far larger than the autumn maximum (October). The lowest number in this year was recorded in September.

In 1979, the growth pattern of P. boryanum was quite different. Spring development of the alga started in February after the ice melted and reached a maximum in late May. The lowest numbers and virtual absence of the alga coincided with August - September.

The winter maximum in 1980 was also in contrast to that of previous years, showing that P. boryanum can also grow well

Fig.25. Seasonal periodicity of Pediastrum boryanum (———) and P. duplex (----).


700
500
250
50
cells / ml -1
(884)
(1191)
(860)
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

during cold months.

The present data in part agrees with HODGETTS (1921) who found Pediastrum to be most abundant in the warmer months, reaching a maximum at the end of the summer or in early autumn with no apparent relation to sunshine hours. The cycle of P. boryanum in Shearwater is also partly in harmony with RAO (1955) who observed Pediastrum to be abundant from April to September but disagrees with SREENIVASEN et al (1964) who recorded Pediastrum to have its peak in January.

The growth pattern of P. duplex was however quite regular, producing small peaks only during autumn and summer. Increasing numbers of P. duplex was generally synchronous with rising populations of P. boryanum. However, the greatest growth of P. duplex coincided with rapidly decreasing numbers of P. boryanum and may indicate the possible competition between these two algae.

The distribution of coenobia with different cell numbers for each year is shown in the following table:-

	8 celled	16 celled	32 celled	64 celled
1977	11	111	77	-
1978	103	350	127	2
1979	29	114	48	4
1980	22	39	37	1

Table 3. Total annual numbers of coenobia of P. boryanum and P. duplex, formed by different numbers of cells.

It is apparent from table 3 that 16 and 32 celled coenobia of Pediastrum spp. were found more frequently while 8 celled ones were less numerous and 64 celled coenobia were extremely rare. Numbers of these different coenobia increased simultaneously with the increasing Pediastrum spp.

The influence of physico-chemical factors on the growth of Pediastrum spp. was complicated in this study. High numbers of the algae were mostly coincident with high temperatures; but the opposite could occur. With regard to the largest maxima, temperatures between 11 and 17°C appeared to be most favourable for growth. However, the lowest numbers and virtual absence of the algae were also recorded at similar temperatures. In addition P. boryanum was also increasing in numbers under the ice in 1980 while it totally disappeared during the ice period in 1979. However, the persistent occurrence of P. boryanum clearly demonstrates high temperature tolerance. P. boryanum was also highly tolerant to chemical changes in Shearwater occurring at different concentrations of dissolved nutrients. Concentration of nitrate and phosphate were usually increasing or already at high levels during periods of active growth of these algae. However, the absence of the algae was also synchronous with increasing or high levels of dissolved nutrients.

Summary and conclusions

Pediastrum boryanum was a conspicuous member of the phytoplankton and was present almost throughout this investigation while P. duplex occurred in small numbers.

Growth of P. boryanum was quite unpredictable while P. duplex showed regular summer - autumn growth.

The algae were represented mainly by 16 and 32 celled coenobia while 8 and 64 celled coenobia were found less frequently.

P. boryanum showed a great tolerance to physico-chemical changes in Shearwater.

Ice formation did not appear to influence the development of P. boryanum.

Scenedesmus quadricauda Chodat

S. quadricauda was another important component of the phytoplankton in Shearwater, occurring on most occasions during this investigation.

The seasonal periodicity of S. quadricauda shown in fig. 26 clearly shows that the distribution was erratic and maxima were attained in every season of the year. However, a winter - spring development of the alga in 1978 and 1980 was more or less similar in terms of numbers and periods, starting in December and terminating by late May. Short growth peaks were characteristic of 1977, otherwise the alga was present in the phytoplankton for long periods and actually disappearing only for short times. The maximum numbers were recorded in July and October 1978, March 1979 and December 1980. The maximum in July 1978 was exceptionally large, consisting of 2711 cells/ml. The cloest number to that was found in October 1978 (327 cells/ml.) otherwise the records were less numerous remaining under 100 cells/ml.

Table 4 illustrates the total numbers of colonies with different number of cells for each year.

	2 celled	4 celled	8 celled
1977	1	30	-
1978	1	956	65
1979	1	96	5
1980	-	147	6

Table 4. Distribution of different cell numbers per colony of Scenedesmus quadricauda.

Fig.26. Seasonal distribution of Scenedesmus quadricauda.

(2711)

100

10

cells / ml^{-1}

A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

1977 1978 1979 1980

It is clear from the above table that the numbers of 4 celled colonies outnumbered 2 and 8 celled colonies in each year. The highest numbers of colonies were synchronous with the largest maximum of S. quadricauda.

Persistent occurrence of S. quadricauda in Shearwater clearly demonstrates that the alga was quite tolerant to changes in physico-chemical conditions. The growth of the alga did not appear to be influenced by temperature since high or small numbers coincided with similar temperatures. Concerning the exceptionally large maximum in July 1978, one might suggest that high temperature favoured its growth. In addition, S. quadricauda was increasing in numbers under the ice when Shearwater was frozen over in 1979 and 1980. Numbers also increased or declined regardless of the fluctuations in concentrations of dissolved nutrients. However increasing or high levels of nitrate and phosphate coincided with the large development of S. quadricauda.

FRITSCH (1903) found Scenedesmus to be common at the beginning of April or a little earlier and RICE (1938b) related the periodicity of S. quadricauda to high sunshine and found it abundant in spring to autumn. RAO (1955) also observed S. quadricauda numbers to be high during spring - autumn but related its periodicity to high temperature rather than sunshine. RODHE (1948) found that S. quadricauda could continue to increase if nitrate or phosphate, or both, were removed from the culture medium by utilizing its own organic nitrogen and phosphorus. The last finding may hold true for the occurrence of S. quadricauda at low concentrations of nitrate and phosphate in Shearwater.

Summary and conclusions

Scenedesmus quadricauda was persistently present in the phytoplankton.

The growth pattern was erratic and maxima were achieved in every season of the year.

The alga was represented mainly by 4 celled colonies.

The growth of the alga did not appear to be influenced by temperature or ice formation and it occurred at low as well as high concentrations of dissolved nitrate and phosphate.

DESMIDIALES

Although few species of the order Desmidiales were found during this investigation they support the eutrophic status of Shearwater.

Staurastrum pingue Teiling and S. longipes (Nordst.) Teiling were the only members of the genus and their total seasonal numbers are plotted together with that of a Closterium sp. (Fig. 27).

The occurrence of Closterium sp. was conspicuous only in 1978 and it was absent in the other years. The numbers of the alga began to increase in November and reached a maximum in March (14 cells/ml.). The decline of the alga was very rapid and few cells were encountered during summer. The alga then disappeared from Shearwater.

Staurastrum spp. were quite persistent occurring on almost every occasion during this study.

The algae were most abundant in 1977 and 1980.

Two maxima were produced by Staurastrum spp. with almost similar numbers; one in autumn and another one in spring. Autumn development commenced in September and the maxima were achieved either in October or in November. The autumn maximum in 1977 was the largest of all. Spring development of Staurastrum spp. begain in late winter and the maxima were attained regularly in May. Although during the summer months Staurastrum spp. were generally sparse, two conspicuous peaks also occurred during summer indicating that they can also develop during the warmest period if the conditions are favourable.

Fig.27. Seasonal variations in the total number of *Staurastrum pingue*, *S. longipes* (_____) and *Closterium* sp. (.....).

(40)

30

20

10

5

cells / ml^{-1}

A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F

1977 1978 1979 1980 1981

Development of Staurastrum spp. was synchronous with either increasing or decreasing water levels and temperatures. However, very low or very high temperatures did not appear to be favourable for their growth. Growth was variable under ice. Autumn and spring developments of Staurastrum coincided with increasing or already high levels of nitrate and phosphate. This may suggest that increasing nitrate and phosphate favour the growth of Staurastrum spp. in this study.

Summary and conclusions

Staurastrum pingue and S. longipes were the only conspicuous members of the Desmidiales, both occurring persistently throughout this investigation.

The desmids, like diatoms, produced autumn and spring maxima regularly. The size of the maxima differed from year to year.

Growth of Staurastrum spp. coincided with increasing as well as decreasing temperatures. Very warm and cold periods were unfavourable for the desmids.

Increasing concentration of nitrate and phosphate appeared to favour their development.

ZOOPLANKTON

The seasonal periodicity of the more common zooplankters in Shearwater is indicated in Fig. 28. It is apparent from the same figure that Amoeba-type organisms were found more frequently occurring in every season of the year.

Keratella cochlearis Gosse generally appeared in small numbers but a sharp increase in numbers was also recorded on one occasion. However, numbers of Amoeba-type organisms and of Trichocerca sp. also increased simultaneously with that of K. cochlearis. Occurrence of Keratella quadrata Müll., Rotifera sp. and Daphinia sp. were almost negligible.

Fig.28. Seasonal distribution of zooplanktoners.

(.....) Keratella cochlearis

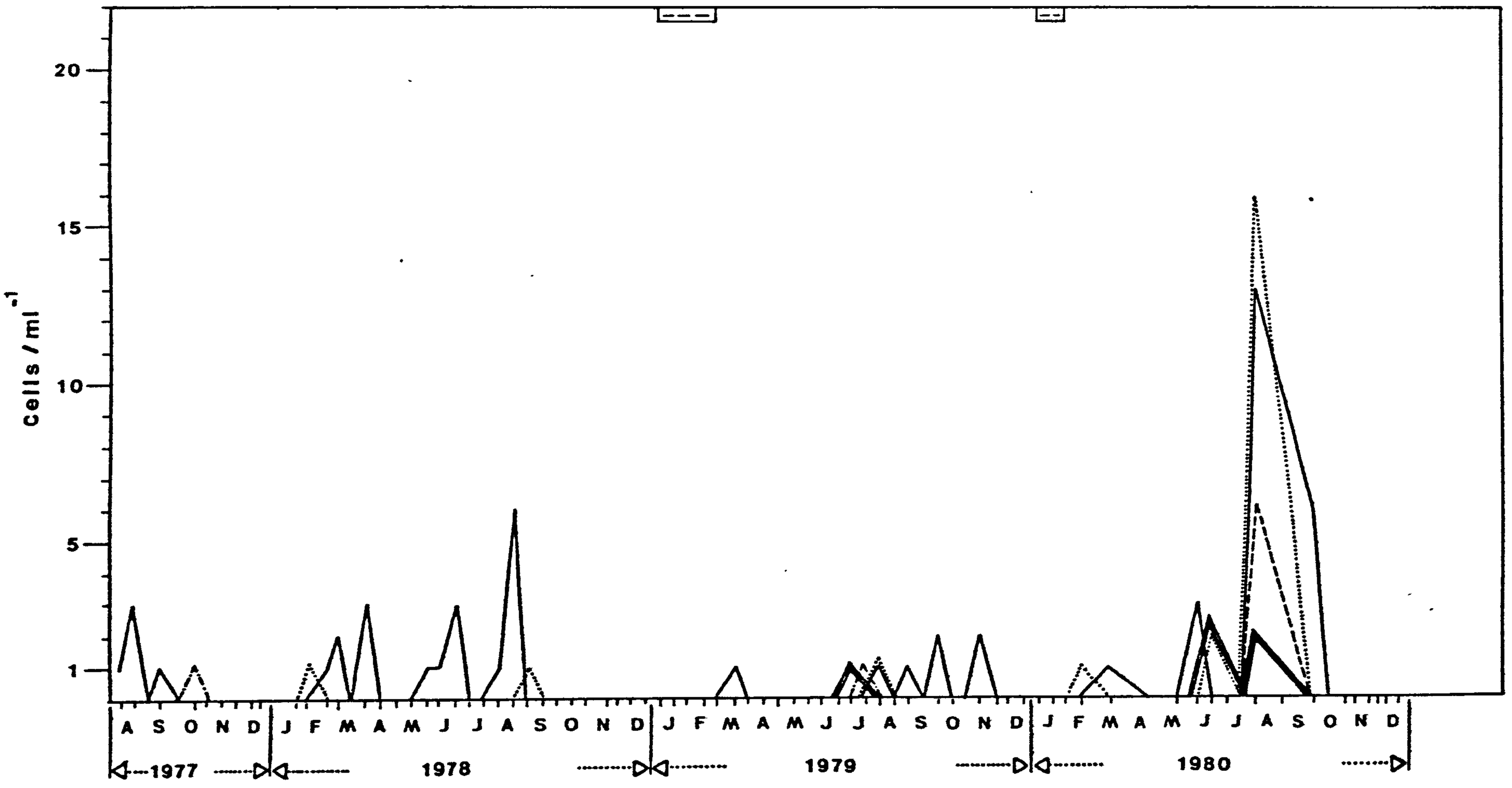
(———) K. quadrata

(- - - - -) Trichocerca sp.

(— — —) Rotifera sp.

(-.-.-.-.-) Daphinia sp.

(———) Annaeboid protozoa


Cells / ml^{-1}
20
15
10
5
1
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1977
1978
1979
1980

CHAPTER 3.

FUNGAL PARASITES OF THE PHYTOPLANKTON

Aquatic fungi together with bacteria and protozoa play a significant role in the nutritional cycle of aquatic ecosystems as they carry out the break-down process of dead organic matter and return the nutrient materials to the environment. In addition, destruction of living photosynthesizers as well as of non-photosynthesizers by aquatic fungi in the aquatic environment is also known with certainty as WESTON pointed out in 1941. WESTON (1940) has also discussed the role of aquatic fungi in hydrobiology suggesting that representatives of phycomycetes are present in all types of inland waters and emphasizes that members of practically all major groups of freshwater organisms are attacked by aquatic fungi.

Although representatives of all fungi are to be found in aquatic habitats, the majority of the fungal parasites and saprophytes associated with phytoplankton are members of the uniflagellate Chytridiales and biflagellate Lagenidiales in both freshwaters and oceans. Both orders belong to the Phycomycetes, which is the most abundant group in freshwaters in terms of numbers of species. However the Ascomycetae infect macroscopic seaweeds. Most of the uniflagellate forms (Chytridiales) are partly or fully external parasites (Epibiontic) while the biflagellate forms (Lagenidiales) are internal parasites (Endobiontic) (LUND, 1957). Some show a degree of host specifity (LUND, 1957).

The Chytridiales, or chytrids as they are commonly called, are not all truly aquatic although they occur predominantly in freshwater - any soil also has a regular and constant flora of chytrids. They differ from all other Phycomycetes in that many are cellulolytic, while others grow on keratinous and chitinous substrata. Their life histories are diverse in detail but in general most show certain features in common. Asexual production is mainly by uni-flagellate spores called zoospores, formed in a sporangium. Zoospores are the main dispersive agents of these fungi and ultimately they find new algal cells upon which they grow. The chytrid zoospore is spherical, small in diameter (circa 2-5μ), contains a single bright refractive globule and has a long flagellum placed posteriorly which propels the zoospore from the rear. In contrast the zoospore of a biflagellate fungus is often bean or grape seed in shape, it has several refractive globules and the two flagella are laterally inserted but oppositely directed when swimming. Many chytrids also produce asexual or sexual thick walled resting spores.

REVIEW OF PREVIOUS WORK

Early observers had often considered the chytridiaceous fungi to be the reproductive structures of the aquatic organisms on which they were living. For example, references are found in the early literature on the formation of zoospores by desmids (ARCHER, 1860), of antherozoids by saprolegnians (PRINGSHEIM, 1860), and of sperms in eggs of the animal Nais (CARTER, 1858). Similar misinterpretations were made regarding marine organisms (WRIGHT, 1879b). As knowledge of BRAUN's monograph (1856a) became more general, however, the extraneous nature of the chytridiaceous fungi was recognized.

In the early years of aquatic research, it was generally assumed that chytrids were the primary causal agents when they were found on dead or dying algal cells. However, ROSEN (1887) soon pointed out the saprophytic tendency of chytrids in connection with a study of Phlyctochytrium zygnema whose zoospores most often came to rest on dead cells of the alga Zygnema or on healthy green filaments which were fast becoming moribund due to unfavourable environmental conditions. Later this view was emphasized by SERBINOW (1907), who studied a number of different chytrid parasites of algae, and concluded that many of them were at most facultative parasites. SERBINOW's view received support from numerous subsequent observations, and it is probable that they hold true for a large number of the Phlyctidiaceae and Chytridiaceae. Certain members of these families appear, however, to be truly parasitic and have never

been induced to live saprophytically (F.K. SPARROW, 1960, aquatic Phycomycetes book). In addition there have also been reported a large number of species, which seem capable of living either as saprophytes or parasites (see, especially SERBINOW, 1907).

Recent studies have also shown that many chytrids apparently have a saprophytic existence. These forms occur wherever suitable substrata are available in the natural habitat.

INGOLD (1944) described a new species Amphicypellus elegans INGOLD as saprophytic when it was found growing on dead thecae of the dinoflagellate Ceratium hirundinella O.F. Müll. PATERSON (1960) and CANTER (1961) have supported the status of saprophysitism for this fungus. PATERSON (1960) studied saprophytism for the first time in quantitative terms in the case of A. elegans on C. hirundinella, and its relation to physical-chemical factors in the environment. No correlation was found between numbers of dead Ceratium cells and times when high numbers of fungal thalli were observed. The fungus did not seem to be affected by varying pH or alkalinity values but by the temperature as three maxima of the fungus occurred within the narrow range 19.2 - 21.5°C. Actual oxygen level differed widely (5.2 - 10.3 ppm) and it is therefore unlikely that falling oxygen levels were important for the development of the fungus.

PONGRATZ (1966) also observed C. hirundinella populations in Lake Geneva over ten years and recorded only one bloom of A. elegans at a time when the surface temperature was only

13.7°C which is much lower than that recorded by PATERSON (1960). The dinoflagellates Peridinium willei (Huitz.) Kaas and P. cinctum Ehberg. were also present in the plankton at that time but no trace of Amphicypellus was observed on these organisms. This might be a good example for the selectivity of this fungus. PONGRATZ, however, called the fungus a parasite and attributed the disorganized appearance of the algal cells to the effect of the fungus.

Yet another fungus Rhizophydium couchii was described as a saprophyte of Pediastrum duplex var. clathratum and var. reticulatum (MASTERS, 1970) since it was observed growing upon coenobia of these green algae which had disorganized cell contents. The greatest numbers of chytrid thalli were recorded while numbers of P. duplex were falling. Temperature appeared to exert a determining effect on the growth of the fungus which was observed most frequently in the temperature range 24 - 25°C, and it did not occur as low as 12.6°C. The importance of pH was less obvious. Pediastrum boryanum was also present in Lake Manitoba, Canada at the same time and in higher numbers than P. duplex varieties but it did not support the growth of R. couchii, however, a polyphagous fungus similar to Podochytrium was occasionally observed in coenobia of this species. This is another good example supporting the concept of host specifity by a fungus.

Chytridium (Diplochytridium) marylandicum Paterson colonizes the alga Botryoccus braunii Kütz. in Delta Marsh, Manitoba when the algal population approaches its maximum although the previously described fungi are usually associated with declining algal populations (MASTERS, 1971c). Neither the alga nor fungus showed

any correlation with pH but was correlated with temperature and conductivity. It is remarkable that C. marylandicum occurs only on healthy Botryococcus colonies although it is described as a saprophyte since it does not exploit cell contents, but only the copious extracellular mucilage around the cells. C. marylandicum appears to be highly specific because it has not been observed on other substrata.

The apparently healthy appearance of an algal host at the time of zoospore encystment and germination usually leads the observer to conclude that the fungus is a parasite. Thus the subsequent unhealthy appearance of the alga would then be due to the fungus.

The bulk of the literature on fungal parasites of algal hosts consists mainly of taxonomic descriptions. Over a century of continuous investigations, particularly in the last four decades, a considerable body of information has accumulated on the diversity of types of aquatic phycomycetes, their structure and life cycles but there has been less attention to their ecology.

The majority of freshwater algal parasites described have been members of the order Chytridiales although a few belong to the order Lagenidiales (simple biflagellate, holocarpic species) (see SPARROW, 1960; KARLING, 1977). However a species of Blastocladiella (order Blastocladiales) was described as parasitic on the blue-green alga Anabaena flos-aquae (Lyngb.) Bréb. (CANTER and WILLOUGHBY, 1964).

Although there have been few investigations in the early years of this century (see de WILDEMAN, 1900, 1931) on the

fungal parasites of planktonic algae, a series of continuous studies (investigations) were started by INGOLD (1941) who described the chytrid Endocoenobium eudorinae Ingold as a parasite of Eudorina elegans. HUBER-PESTALOZZI (1944) described a new species Chytridium oocystidis parasitizing Oocystis lacustris Chod. and he also figured (1946) chytrids on Asterionella formosa Hass., Sphaerocystis schroeteri Chod. and on Fragilaria capucina from Switzerland. From Sweden descriptions of many parasites of algae are made by SKUJA (1948). He recorded Olpidium entophytum (A. Braun) Rabenhorst in both Gloeocystis bacillus and G. planktonica; another chytrid O. endogenum Braun occurs in the desmid Cosmarium depressum var. achondrum. These descriptions by SKUJA received criticism from CANTER (1950) as she found a very similar parasite occurring in Gloeocystis in the English Lake District. This parasite produced biflagellate zoospores. SKUJA did not observe the zoospores in his form and it is possible, especially from his drawing (1948, taf.49, fig.7), that this species is also a biflagellate and does not belong to the genus Olpidium. SKUJA also described a new chytrid Phlyctidium anabaena Rödhe & Skuja which is parasitic in young resting spores of Anabaena spp., and Chytridium microcystidis Skuja, a parasite of Microcystis spp. in Erken. Remaining records from Sweden belong to the diatom Melosira which was parasitized by Rhizophydium simplex (Dang.) Fischer, R. fusus (Zopf) Fischer and by Chytridium versatile Scherfell.

After SKUJA, parasites of planktonic blue-green algae were investigated in the English Lake District. Rhizosiphon crassum

was observed attacking Anabaena solitaria and other Anabaena spp. while the closely related chytrid R. anabaenae which was originally described as Phlyctidium anabaena by Rodhe and Skuja (SKUJA, 1948), attacked Anabaena sphaerica, A. spiroides and A. macrospora (CANTER, 1951). Blastocladiella anabaenae which may belong to the Blastocladiales (CANTER & LUND, 1968) occurred predominantly on Anabaena flos-aquae as well as on Aphanizomenon flos-aquae, Anabaena circinalis and A. solitaria.

Many chytrid parasites of planktonic blue-green algae appear to be confined to a specific structure such as a resting spore or heterocyst: Rhizosiphon akinetum apparently attacks only the akinetes of Anabaena affinis and of A. macrospora (CANTER, 1954) while Chytridium cornotum is found only on the heterocysts of Aphanizomenon (CANTER, 1963).

Most of the planktonic algae in freshwaters have been found to be susceptible to fungal parasites but the effect of the parasitism on natural populations has rarely been recorded. By the beginning of this century writing on the role of chytridiaceous fungi which infest phytoplankters WESENBERG-LUND (1905) stated the following "I have shown, further, that nearly all the protoplasm of the cells in the plankton is eaten by phycomycetes before reaching the bottom: my observations prove that an organism in the latter part of the period of maximum development may very often be infected by phycomycetes, which feed upon the protoplasm and kill it leaving the skeleton intact". This statement somehow emphasizes the fungal effect on the host in terms of feeding but gives no evidence of fungal effect on the growth of the host. REYNOLDS (1940), however,

showed that cell numbers of a form of Staurastrum paradoxum Meyen were reduced by a chytridiaceous fungus. In addition, the role of aquatic fungi in hydrobiology has been reviewed by WESTON (1941, esp.p.142).stressing the probable importance of parasites in controlling numbers of planktonic algae.

First critical investigations on the effect of parasitic fungi in relation to fluctuations in the numbers of planktonic algae in freshwater seem to have been commenced by the studies of CANTER and CANTER & LUND in England and elsewhere,and these have continued over more than thirty years.

In recent years CANTER (1946 - 1969) and CANTER & LUND (1948, 1951, 1953) have gathered a great deal of information on English Lake District waters (four bodies of water: Windermere, North and South Basin, Blelham Tarn and Esthwaite Water) concerning the parasites and saprophytes of planktonic algae, and their influence on the phytoplankton population cycle. They found many chytrids of various genera occurring in significant numbers on phytoplankters. In general their study consists of detailed analyses of the incidence of parasitism on phytoplanktonic algae, the influence of the fungi in reducing phytoplankton populations and the interaction between parasitism and other factors determining the growth of algae. Their intensive works present a clear demonstration of fungi at work in lakes parasitizing primary producers. The majority of the larger planktonic algae of the English Lake District belonging to the classes Chlorophyceae, Bacillariophyceae, Chrysophyceae, Xanthophyceae, Myxophyceae, Dinophyceae and

Cryptomonadineae; all are parasitized from time to time by one or more fungi (mainly belonging to the order Chytridiales and a few belong to the Lagenidiales). They recorded about fifty different species of phycomycetes - three saprophytic, the rest parasitic - on the planktonic algae. Some of these fungi were found to be infected by hyper parsites (e.g. Rozella parvum in Zygorhizidium affluens) (CANTER, 1969). In addition CANTER described over ten new species of phycomycetes on algae.

Their quantitative limnologial studies of chytrids on planktonic algae were pioneering works, laying the foundations with which subsequent data can be compared (CANTER & LUND, 1948, 1951, 1953, 1969). Their careful quantitative works revealed several interesting situations with regard to the relationship between fungal parasitism and other factors acting on diatoms and other algae (e.g. desmids). One situation was concerned with the effect of parasitism on algal maxima. CANTER & LUND (1948, 1951, 1953) studied the parasitism of the chytrid Rhizophydium planktonicum Canter Emend (renamed in part Zygorhizidium affluens Canter 1969) on the diatom Asterionella formosa Hass over a long period. They (1951) proposed that the parasites may delay the time of the algal maximum or may decrease the size of the algal maximum if the parasite reaches epidemic proportions. In addition the parasitism of one alga may favour the development of other (fungi-resistant) algae. However, CANTER & LUND (1948); LUND (1949, 1950) also suggest that the parasitism is only one of the factors controlling the changes

in density of the population from time to time and physical-chemical factors operate at other times. The possibility of forecasting approximately when a particular fungus will occur in any lake has also been put forward by CANTER (1954) since several of the common fungi occur at almost the same time of the year and an arbitrary lower limit for epidemics has been set by LUND (1957) at one quarter infection of the host population.

More recently a quantitative study of desmids infected by fungi (CANTER & LUND, 1969) once again demonstrated the dramatic effects of parasitism on healthy species of desmids by reducing dramatically the numbers of the algal population. Although fungal epidemics may be severe, however, they do not alter the normal seasonal periodicity of the desmids. In conclusion these authors do stress that many species of desmids would be more abundant in Windermere and other lakes in the absence of the chytrid and biflagellate parasites. They noted that parasites of desmids were more often biflagellate (Lagenidiales) than chytrids.

Although CANTER and LUND pose many unsolved problems, they do give us a new approach to the study of a chytrid epidemic, and demonstrate beyond question the importance of parasitism in limnology and algal ecology. However their results are for large, only slightly eutrophic waters and we cannot expect that all aspects will be comparable with Shearwater which is a small lowland moderately eutrophic lake.

Other recent studies concerned with the mycology of

planktonic fungi have been made by SPARROW (1951), and PATERSON (1956, 1958) and limnological investigations have been published by PATERSON (1957, 1960) in the U.S.A.

PATERSON (1960) studied the degree of parasitism caused by Rhizosiphon anabaenae on the blue-green alga Anabaena planktonicumin relation to physical-chemical factors which seemed to influence the number of parasites. Dissolved oxygen level was found to be the limiting factor for the parasite as numbers of Rhizosiphon decreased sharply at high dissolved oxygen level, decrease in pH and carbonate alkalinity was also suggested by the author as other possible factors in the occurrence of parasitism. He found the temperature to have little direct effect on Rhizosiphon maxima thus confirming temperature studies by CANTER and LUND (1948, 1951, 1953).

PATERSON (1967) also studied the vertical distribution (0-80m) of chytridiaceous fungi attacking planktonic algae in Grand Traverse Bay, Lake Michigan. Infested algae consisted of only diatoms although other algaewere present in the samples (e.g. Pediastrum, Dinobryon, Ceratium, etc.) He found that numbers of diatoms of all forms was greater at 25 and 30m and there supported parasitic growths. PATERSON thinks that the distribution of fungal populations may have been influenced by the thermal stratification of the water. This is in harmony with LUND's (1957) suggestion that thermal stratification can influence the distribution of the host therefore limiting the invasion by chytrid zoospores.

Apart from the influence of temperature, PATERSON found very little relationship between physical-chemical factors and the occurrence of fungi on diatoms since dissolved oxygen, total alkalinity and pH were quite uniform from the surface to 75 cm.

PONGRATZ (1966) studied the effects of fungal parasites on algae of Lake Geneva, Switzerland for ten years. Although his observations were interesting they were lacking in quantitative data. Among other algae, he noted a pelagic filamentous green alga Mougeotia gracillima (Hass.) Wittrock present throughout the year and capable of producing blooms from early summer to late autumn. Attacks of a Rhizophydium (he named it R. mougeotia) species were observed in the cold months while the alga was still clearly dominant in the plankton. Within a month the number of algæ decreased greatly and subsequently only limited numbers were observed.

A study of parasitism of green algae was undertaken by FOTT (1967), who described Phlyctidium scenedesmi Fott as a virulent parasite on Scenedesmus quadricauda from Czechoslovakia. The fungus was observed during a cold, rainy summer and FOTT suggested that such weather was an important factor favouring attack of the fungus. However this suggestion has been discounted by cultural studies of Phlyctidium scenedesmi var. acuti on Scenedesmus acutus f. alternans and on S. armatus (SOEDER & MAIWEG, 1969) and also by the ecological studies of P. scenedesmi on Scenedesmus quadricauda and Pediastrum boryanum (MASTERS, 1971d). These authors stated that high temperatures tend to favour the

growth of the chytrids. Their data also show that parasites play a central role in controlling the cycle of algal populations, since they reduce the cell numbers of the dominant species under severe attack and give a chance to otherwise suppressed species to develop. These shifts continued from dominant to suppressed until the chytrids disappeared completely (SOEDER and MAIWEG had to use fungicide). MASTERS (1971d) admitted that the factors that favoured the growth of Phlyctidium scenedesmi were unknown as the first summer was warm and the next one was cold and rainy. When Pediastrum and Scenedesmus populations were found to be low in numbers, the infection was also scarce (cf. the work of LUND & CANTER (1948) showing that algal cell numbers have to reach a certain level before an epidemic can occur). Moreover none of the environmental parameters studied, including pH, temperature and conductivity, appeared to be correlated with the fungal epidemics or with algal maxima.

As new papers continue to appear, it becomes apparent that these parasites can occur wherever suitable algal hosts are found. GEITLER (1965), for example, described Dangeardia sporapiculata var. minor Geitler which attacked the alga Heleocharis pallida Korsikov growing on constantly moist wood of farm buildings in the eastern Alps. In addition STEIN and AMUNDSEN (1967) and KOL (1970) recorded chytrid sporangia on Chlamydomonas cells in red snow.

Several of the recently described fungi are parasites of motile algae, most of these fungi, however, attack the algae while the host is at immotile stage. GEITLER (1962) observed the attack of Dangeardia sporapiculata Geitler on Chlamydomonas

which were in the "palmella" stage while Rhizophydium fugax Canter (CANTER, 1968a) attacks Cryptomonas during its quiescent stage, R. nobile Canter (CANTER, 1968b) parasitizes resting spores of Ceratium hirundinella O.F. Müll, and Pseudopileum unum Canter (CANTER, 1963) attacks cysts of Mallomonas.

Recent studies have also proved most interesting in that different growth forms of hosts exhibit a different degree of susceptibility to fungal parasitism. KOOB (1966) for example, separated five distinct populations of Asterionella formosa based on the frustule lengths. It was very interesting that only one population (β population) was attacked by Rhizophydium planktonicum Canter and other populations occurring at the same time remained fungus-free.

The pattern of attack by Phlyctidium bumilleriae Couch on Staurastrum pinque Teiling was full of paradoxes (MASTERS, 1971e). Staurastrum pinque was present in the greatest numbers in the phytoplabkton during the summers of 1965 - 1968, occurring in three different growth forms (three-radiate, four-radiate, and an intermediate growth form with three radii on one semicell and four on the other) but the ratio of the infection by the chytrid on each form varied greatly. In the first two years the four-radiate form was more heavily attacked than the three-radiate form, although the latter was present in greater numbers. The following year, it was the four-radiate form which was more numerous but still most heavily parasitized by the chytrid. Infection of the intermediate form was similar to that of the four-radiate form.

Fungal parasitism of the genus Oocystis was investigated by MASTERS (1971a, 1971b) in the Delta Marsh, Manitoba and in Lake Manitoba, Canada. He described a new chytrid species Chytridium (Diplochytridium) deltanum Masters (MASTERS, 1971b) which was the most conspicuous fungus of all in this lake and attacked several Oocystis spp. simultaneously but the level of attack on different hosts varied considerably (MASTERS, 1971a). However, numbers of the genus Oocystis were low compared with the dominants (e.g. diatoms). Five species of Oocystis were present but Oocystis crassa Wittrock and O. lacustris Chodat were by far the most successful. Heavy attacks of Chytridium deltanum on growing populations of these algae appeared to suppress their maxima. In addition, slight infections of C. oocystidis Huber-Pestalozzi (1944) and a polyphagous inter-biotic parasite were also recorded on these hosts. Rare attacks of a Lagenidium sp. on Oocystis parva West and West and O. submarina Lagerheim occurred occasionally but to a lesser degree. Of the environmental parameters examined, for correlation with the onset of the fungal attacks on Oocystis species, only temperature appeared to be in any way a contributing factor.

CULTURE WORK

Culture work is very essential to determine the precise taxonomy, life cycles of fungal parasites and saprophytes, and their effects on the algal hosts.

Experimental growth studies of chytrids and other fungi, in clonal cultures, on both live and dead algae have been the subject of several papers in recent years. The influence of different

hosts as well as the density of cells of the host on the morphological variations of the developing chytrids were recorded by KOCH (1957); REGISTER (1959); PATERSON (1963); JOHNS (1964); BARR and HICKMAN (1967). The physiological properties of the developing chytrid were studied by KOCH (1968); BERNSTEIN (1968); BOTSICK (1968); BARR (1969, 1970a). BOTSICK (1968) also studied the effects of temperature which influenced the morphological size of Chytriomyces hylinus. These studies showed that the developing chytrid displayed different morphological and physiological variations in response to different substrata. The importance of these findings in Chytrid taxonomy was discussed by MILLER (1968).

More recently BARR (1970b) investigated the nutritional requirements of two isolates of Rhizophydium sphaerocarpum. The differing ability of isolates is illustrated by the fact that one grew on ten different boiled substrata whilst another completely failed to grow on these substrata although it did grow on nutrient media in pure culture. In conclusion he emphasized the importance of nutritional requirements and their influence on the variation of morphological characteristics of chytrids. Studies of HASIJA and MILLER (1971) and MILLER et al (1973) supported this view. In addition morphological and developmental studies of single-spore isolates of Entophylyctis (Chytridiales)(BOOTH, 1971 and BARR, 1971) indicated the importance of this kind of experimental work for the precise taxonomy of the species.

The major impediment to determination of the factors favouring the onset of parasitism, and their influence on

algal hosts has been the lack of a continuing source of host material owing to the difficulty of culturing many species. Before the work of JOHNS (1960) with Polyphagus there had been no reports of success in maintaining parasites on algae grown in culture. Later however followed the study of COOK (1963), who isolated ten strains of several aquatic phycomycetes (representatives of Entophlyctis, Mitochytridium (Chytridiales), Mezocytium (Lagenidiales) and Ancylistes (Entomophthorales)) parasitic on desmids (e.g. Closterium, Micrasterias) and these he maintained successfully on hosts grown in unialgal culture. In some instances, the life cycles of the parasites were influenced by the hosts and their ability to attack only certain clones was correlated with morphological differences between clones assigned to varieties.

Although a wide variety of algae were used in previous studies none were diatoms. FRIEDMANN (1952) described fungal parasites growing on benthic diatoms in enrichment cultures but there appears to be no report of a fungus being maintained in clonal culture on a freshwater planktonic diatom. Attempts of CANTER and LUND (1948) and PATERSON (1967) to cultivate chytrid parasites on freshwater planktonic diatoms were unsuccessful. However, de WILDEMAN (1900, 1931) stated that cultivation of Rhizophydium schroeteri de Wildeman - a parasite of Asterionella formosa - was easy, but gave no evidence whether or not he prepared any cultures of Asterionella which were subsequently inoculated with the chytrid.

More recently successful isolation and maintenance in clone culture of the chytrid Rhizophydium planktonicum Canter emend.

parasitic on Asterionella was achieved and the ability of this fungal isolate to infect other clones of Asterionella, Fragilaria, Tabellaria, Synedra and Cyclotella as well as dead material were tested by CANTER & JAWORSKI (1978). They noted that all the clones of Asterionella were highly susceptible to infection, whereas there was hardly any infection on other diatoms. They found no evidence to suggest that the chytrid could complete its life cycle on dead material.

Hypersensitive reactions to fungal parasitism in higher plant cells are already well documented and known to occur in a wide variety of host-pathogen combinations (KARLING, 1964; WOOD, 1967, 1972; WEBSTER, 1970). CANTER & JAWORSKI (1979) also stated that Asterionella formosa may undergo a hypersensitive death reaction in response to Rhizophydium infection in culture. In addition they suggested that the light intensity might play some role in the eventual adherence of a zoospore upon a cell of Asterionella. Later sensitivity of zoospores of R. planktonicum to critical light intensities was demonstrated by the same authors (1980, 1981).

Summary of Previous Works:-

It is very obvious from the studies mentioned above that factors affecting the numbers of parasitic and saprophytic fungi on algae are quite confusing. Among all the physical-chemical factors, only temperature seems to have an important limiting and steady effect on these fungi. Even so it is still disputable whether or not the same fungus occurs either at low or high temperatures in different years since different strains of the

species may be involved. Moreover, it has also been suggested (CANTER & LUND, 1948, 1951, 1953; PATERSON, 1960) that temperature appears to exert little direct effect on maxima of parasites. Dissolved oxygen and pH levels tend to have a less important influence on the growth of these fungi. One must also conclude that the host itself does have a great effect on the occurrence of fungi. Two factors are involved. (1) A different growth form of the same host has a different degree of suceptibility to certain fungal parasites, (2) Different hosts as well as the density of the host have an influence on the morphological variation of fungi.

It has now become very obvious that fungal parasites reduce the cell numbers of host populations but it is still difficult to correlate the interactions between parasitism and the physical-chemical factors determining the growth of algae in plankton. Suggestions by different authors are in conflict, for example, one (CANTER & LUND, 1951) points out that the epidemic of parasitism may hasten the algal decline. On the other hand, another author (PATERSON, 1960) insists that the decline is caused by physical-chemical factors, not by parasitism. Moreover, algal pathology is becoming very complex in that not only are aquatic fungi capable of attacking and killing algal cells, but also certain dinoflagellates give rise to detrimental effects on algae (see sp. CACHON et al. 1969 and TAYLOR, 1968).

Very few investigations have so far been made on the effects of parasitic fungi on phytoplankton populations

although it was emphasized and demonstrated (see CANTER & LUND, 1948) that these fungi could exert an important influence on phytoplankton population. For many years in the study of lakes, the physico-chemical factors were considered the main reason for alterations in the numbers of the phytoplankton. The importance of a biological factor such as parasitism was neglected. As so little progress had been made, CANTER & LUND, (1970) wondered "Is Windermere peculiar?" since the majority of larger planktonic algae were parasitized from time to time by one or more fungi.

However, it has recently been shown that dominance can be modified by the effect of attacks of fungal parasities (REYNOLDS, 1973). In addition factors such as silica limitation, fungal parasitism and grazing by a protozoan were suggested as particularly important factors affecting the rise and fall of a diatom population (BAILEY-WATTS & LUND, 1973).

Aim of the Present Work:-

It is certain that the influence of parasitism and physical-chemical properties on the cycle of the phytoplankton population is complex and requires further study. It is therefore the aim of this study to present more data on these subjects.

For this purpose the parasitism and its effect on members of the phytoplankton in Shearwater has been studied for three years (1978 - 1981) in relation to physical-chemical factors. Over the period of study a number of members of phytoplankton

were repeatedly infected by chytrid parasites.

In ecological work observation of the natural population must precede an experimental approach in the elucidation of problems which become apparent. It would have been desirable to do some experimental work on these parasites with algal hosts but as these studies require a considerable amount of time and should continue for a long duration, it must be considered a future project.

There are few detailed quantitative ecological studies of fungal parasitism in the literature and an attempt has been made in this work to compare them with the present study. Finally the role of the parasitism in algal ecology is discussed and suggestions made for further development of the subject. Fungal parasite of each alga are described separately. For the identification of the chytrids CANTER's descriptions are used.

METHODS

Surface samples for algal, chytrid counting and for chemical data were taken at fortnightly intervals. Samples were taken in the mornings (10.30 - 11.00 a.m.). Algal numbers were obtained by sedimentation after addition of a saturated iodine solution to the counting chamber and counting on an inverted microscope by the UTERMÖHL (1931) technique. Infection on the algal cells was determined on fresh samples collected by net hauls. Counts of sixty colonies (sixty cells for single celled species) or more in the case of Asterionella formosa (usually 100 colonies when it was abundant) were made.

Total number of zoospores, living sporangia, empty sporangia and resting spores were analysed separately beside determining the number of parasitized and living cells in each colony. The number of live and dead cells were also recorded. The cells were considered dead either when the cell was empty or chromatophores had completely lost their morphological characteristics. An ocular micrometer was used to ascertain the length of individual frustules of Asterionella. While determining the infection on the cells, the length of frustule was also measured for both infected and uninfected cells. In the case of colonies the length of one frustule was measured. In samples where cells of the colonies had separated measurement was carried out on single frustules. All the drawings of the various stages of the chytrids were made by the aid of Lucida Camera.

Fungal Parasitism of Asterionella formosa by Zygorhizidium affluens Canter

Parasite infection was the heaviest on the diatom Asterionella formosa out of all infections recorded on planktonic algae. Because of this, priority was given to this diatom in the fungal/algal studies.

Chytrid parasitism of Asterionella formosa in relation to its effect on the host population has been studied by few authors (CANTER & LUND, 1948, 1951, 1953; KOOB, 1966; REYNOLDS, 1973 and YOUNGMAN et al. 1976).

CANTER & LUND (1948) described a new chytrid Rhizophydium planktonicum Canter heavily parasitizing Asterionella in the English Lake District. Later it was suggested that R. planktonicum probably represented an aggregate species (CANTER, 1953, 1955 and CANTER & LUND, 1953). Eventually it was confirmed that Asterionella is parasitized by two morphologically very similar species of chytrids, an operculate species was named as Zygorhizidium affluens Canter (CANTER, 1969) whilst the other remained as R. planktonicum. Z. affluens was the one which caused epidemics on Asterionella formosa in Windermere. However, at least six chytridiaceous fungi have been recorded so far on Asterionella in various lakes of the world and some of these have also been found on other planktonic algae.

In the present study, cells of Asterionella were infected only by Zygorhizidium affluens.

Description of Zygorhizidium affluens

Full description of this fungus is given by CANTER & LUND (1948) and CANTER (1969). Therefore a brief description will be given in this section. Life stages in the development of Z. affluens are shown in Figs. 29 and 30.

Thallus monocentric, eucarpic, sporangium develops by direct enlargement of the zoospore. Mature sporangia are usually spherical, 5 - 11μ in diameter (Fig. 29h), occasionally slightly oval (Fig. 29g). Number of oil globules in sporangium varies between 4 - 18 according to the size of the

Fig.29. Development phases of the chytrid Zygorhizidium affluens Canter on Asterionella formosa.

a encysted zoospores

b - c germinating zoospores

d - f developing sporangia

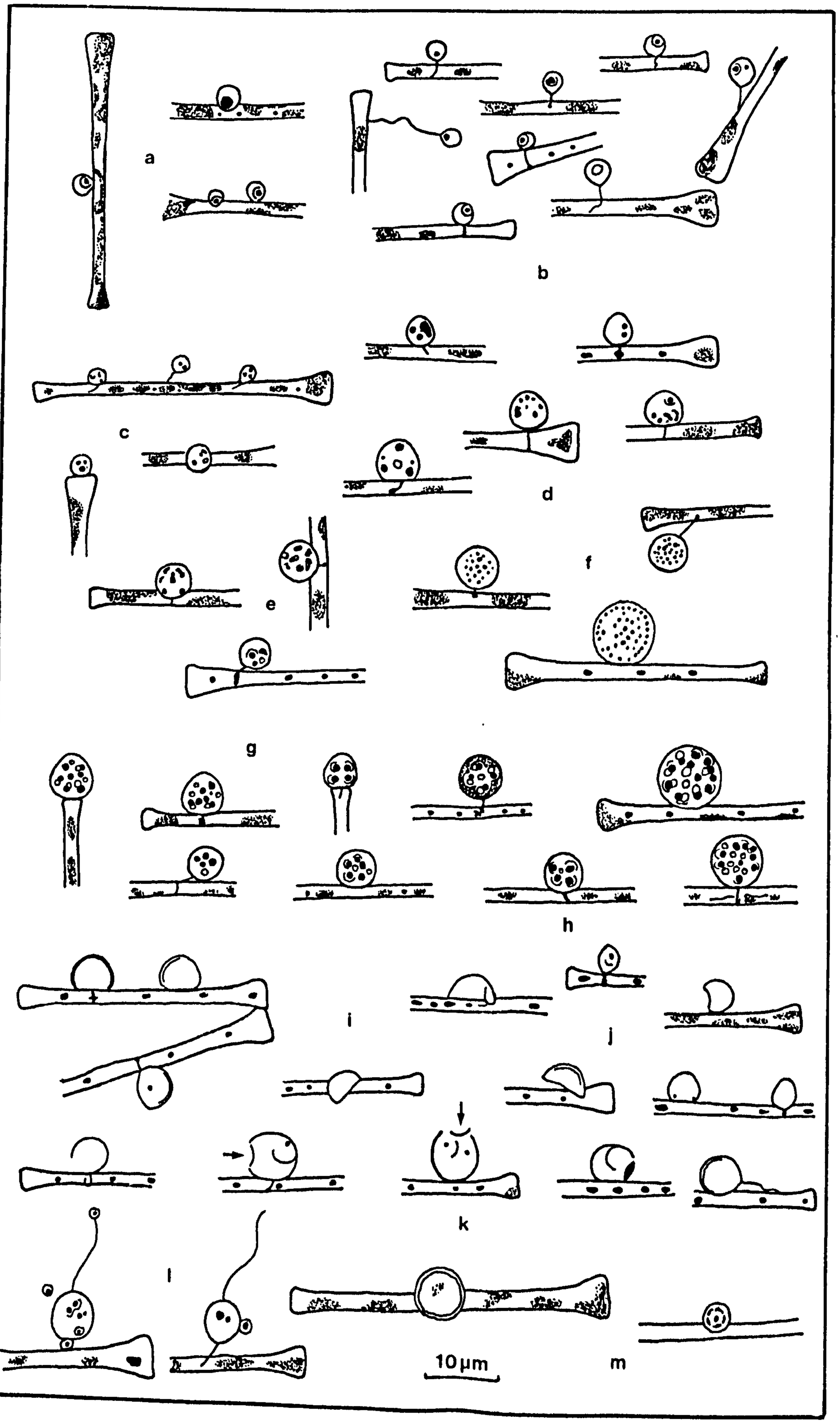
g - h mature sporangia

i - k empty sporangia (→) indicates the lid

l resting spore formation (male cell with a long flagellum)

m resting spores

all pictures at X450


a
b
c
d
e
f
g
h
i
j
k
l
m
10µm

sporangium, each indicating the position of a zoospore. The rhizoidal system (Fig 29b,q) consists of a thicker elongate thread, sometimes with a single lateral branch (Fig. 29b). The elongate thread is smaller in length. Dehiscence mostly lateral (Fig.30p,r,s,t) sometimes apical (Fig.29k). On dehiscence an area of the sporangium wall is rejected as a lid (Fig 29k). Even in small sporangia a lid is present (Fig 30s). In some instances an apical pore is found, developing outwardly (Fig. 29k) through which the zoospores emerge. The sporangium does not collapse after dehiscence.

The zoospore is spherical, 3 - 3.5μ in diameter with a long posterior flagellum (Fig.29b). It contains a single globule: a nuclear cap is visible (Fig 29a).

Resting spores (Fig 29m) are formed sexually after a fusion of a smaller male cell with a larger female cell. The male cell (Fig.291) (essentially an encysted zoospore) is attached to the female cell (Fig. 291). Resting spores are spherical, 5 - 8μ in diameter with slightly wrinkled walls.

Parasitism

Parasitic occurrence of Z. affluens was quite obvious in the present data since the fungus always occurred on healthy cells and on actively growing populations of A. formosa.

Healthy cells of Asterionella have well-developed chromotophores, occupying a considerable proportion of the internal surface of the cell (Fig 30a). Under favourable

Fig.30. Micrographs of developmental stages of Zygorhizidium affluens Canter.

a - b encysted zoospores

c - d germinating zoospores

e - i immature sporangia

j - n mature sporangia

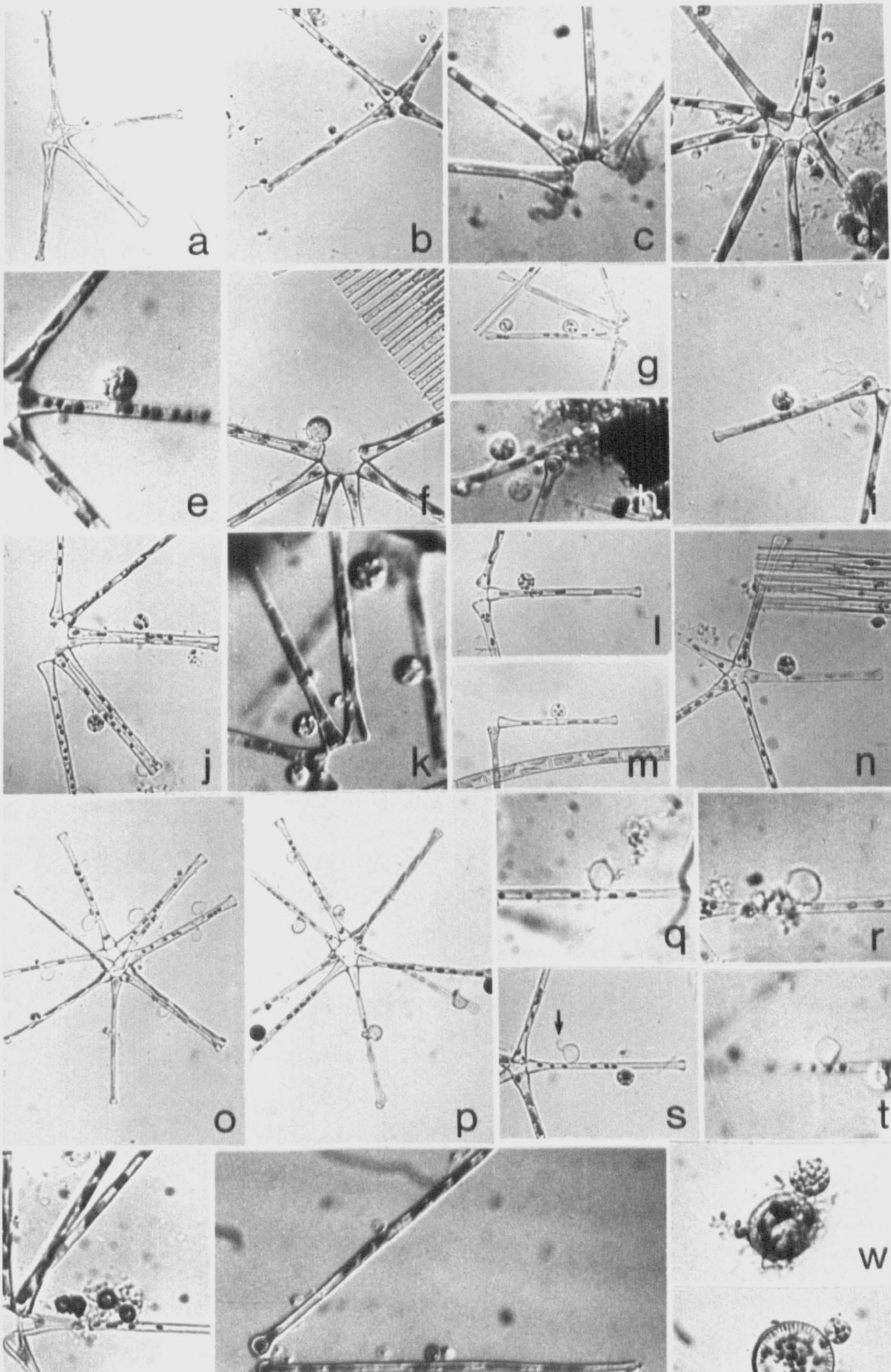
o - t empty sporangia

u resting spores

v encysted zoospores on macro populations of Asterionella formosa

w - x mature sporangia on Cyclotella sp.

all pictures at X410


a
b
c
d
e
f
g
h
i
j
k
l
m
n
o
p
q
r
s
t
u
v
w
x

conditions, the zoospores come to settle down on the host. The zoospores fix themselves to the mucilage or the cell wall of alga. A fine thread (germ tube) is then produced by the encysted zoospore which penetrates the algal cell and forms a rhizoidal system (Fig. 29b). Nourishment is taken into the zoospore by means of a rhizoidal system. The zoospore then enlarges and becomes a sporangium (Fig 29c,d,e). At the beginning of the epidemic the number of attached encysted zoospores is predominant (Fig.30d,v). Single zoospores are always found on healthy Asterionella cells (Figs 29a,) 30a). This is because the attack on the protoplasm of the host has just started and the chromotophores are as yet little disorganized. At the end of active fungal multiplication, numbers of encysted zoospores decrease whilst the numbers of sporangia continues to increase (Fig.31). It is noteworthy however that zoospores are present on Asterionella until the end of the epidemic (Fig. 31). In the last stage of the epidemic, the empty sporangia reach their maximum development in terms of numbers (Fig. 31). It is obvious from this figure that numbers of developing sporangia remained low compared with numbers of zoospores and of sporangia.

Resting spore formation was observed only once and this was during the most severe epidemic(in 1979). They were found after the maximum of the epidemic (Fig. 31). Thus their appearance seems to correspond with high fungal activity. The factors governing the occurrence of resting spores remain obscure in the present study, since they were not observed during other epidemics.

Fig.31. Distribution of developmental stages of Z. affluens Canter during fungal epidemics on A. formosa.


encysted zoospore


developing sporangium

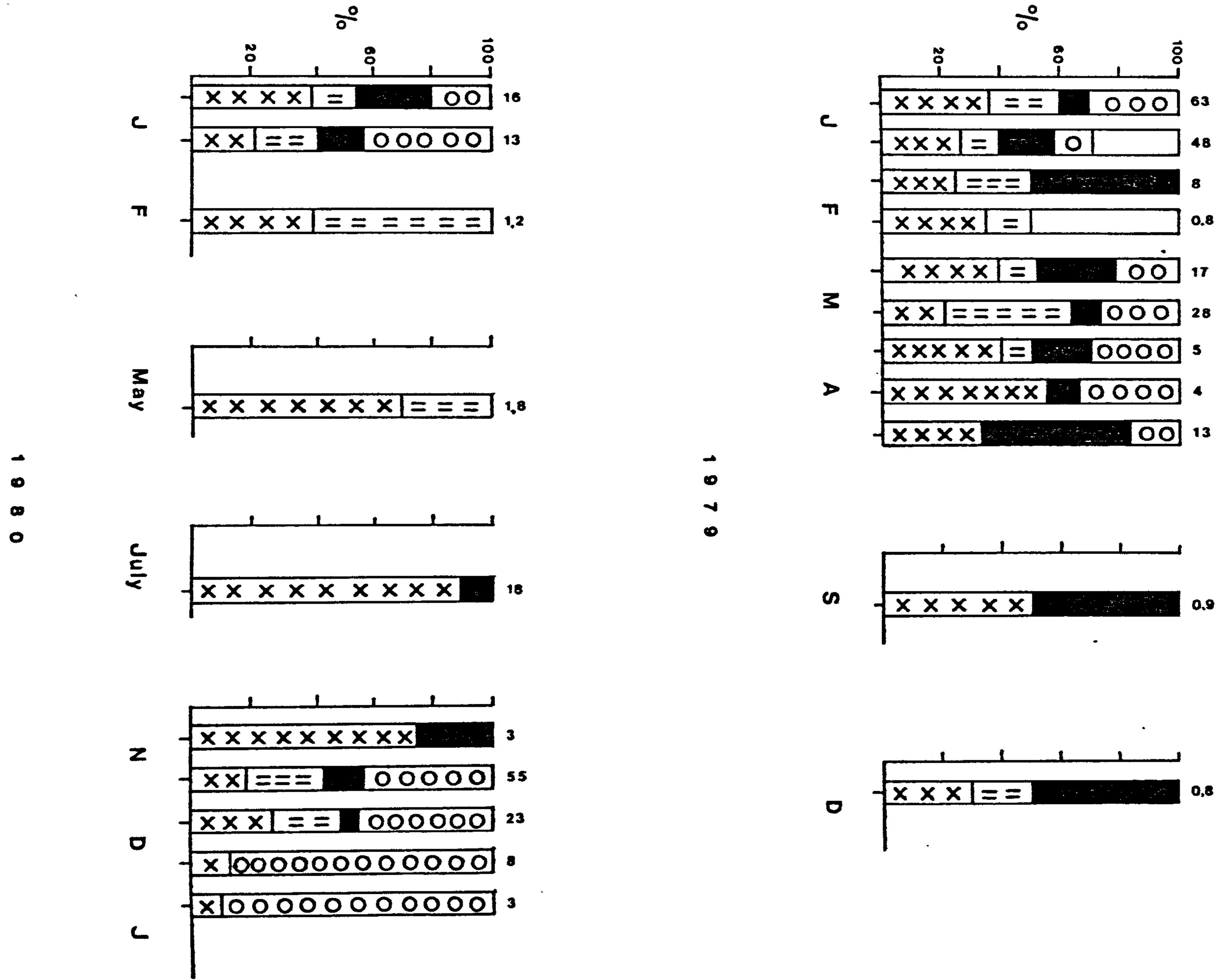

mature sporangium


empty sporangium


resting spore

The same symbols are also used for the distribution of live stages of other chytrids during their occurrence on other planktonic algae.

Note: Above numbers indicate the percentage of fungal infection.


1979
%
20
60
100
J
63
48
F
8
0.8
M
17
28
A
5
4
13
S
0.9
O
0.8
1980
May
1,8
July
18
N
3
55
D
23
8
J
3

During the epidemics the cells bearing well-developed or empty sporangia and resting spores, had either considerably disorganized chromotophores or were wholly destroyed (Figs 29i,t;30^{o-u}). In contrast it was the healthy cells which most frequently bore the zoospores (Fig.29a, 30a-d). HUBER-PESTALOZZI (1946) stated that the cell walls of Asterionella may be deformed as a result of parasitism by chytridiaceous fungi. This was never observed during any epidemics. However, deformed walls did occur with or without presence of Z. affluens. In conclusion the present data agrees with CANTER & LUND (1948) that the fungus population passes through a series of characteristic phases during an epidemic (Fig . 31): the term epidemic refers to the occasional outbreak of severe parasitism. In the present study all the life stages of the parasite are observed and drawn during an epidemic.

Epidemics - in relation to fluctuations in the numbers of A. formosa.

Periodicity of A. formosa in relation to physical-chemical factors was discussed in detail in the previous chapter from a study of the literature. Therefore here it will be mentioned very briefly.

Asterionella was a very conspicuous member of the phytoplankton in Shearwater (Fig 5). Vernal maxima occurred every year whilst much smaller autumn and summer maxima were sporadic in occurrence. Asterionella was present in the plankton

on most occasions and the growth of the alga was interrupted by epidemics of Z. affluens Canter (Fig. 33). Occurrence of epidemics of the fungus varied from year to year. Periodicity of the epidemics of Z. affluens with infection rates and the fluctuations in the numbers of Asterionella are shown in Fig. 32 and in Fig. 33 respectively.

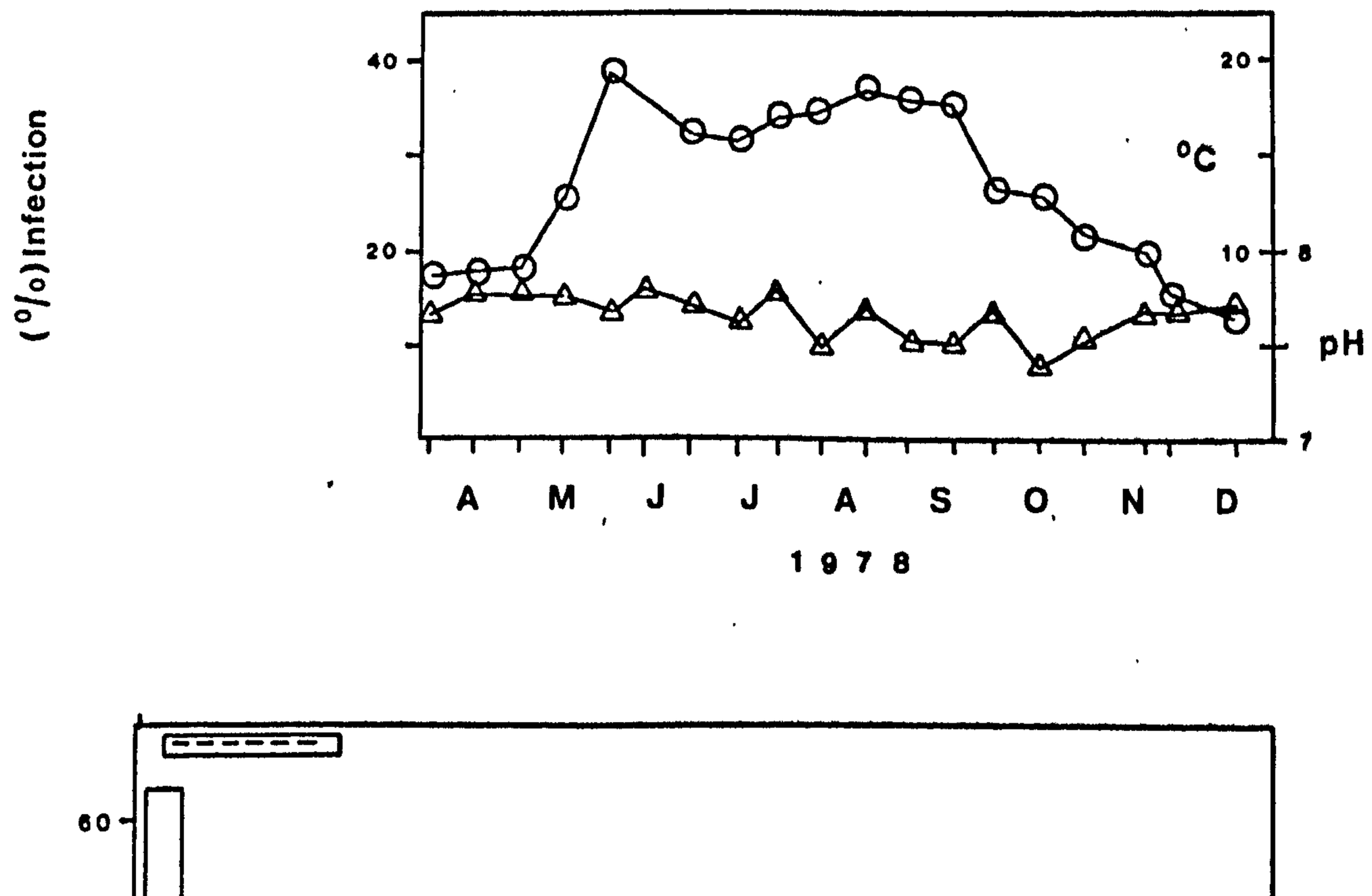
In 1978, from April to the end of December, there was no sign of the chytrid in the samples. The onset of the vernal maximum of Asterionella started in November and numbers reached 42 cells/ml by the middle of December. Numbers of the alga were 383 cells/ml on 9th January (1979) when the first appearance of the chytrid was observed. This infection was exceptional in that a large number of Asterionella were infected (63%). In fact this was the maximum of the epidemic which occurred very quickly without a previous sign. A fortnight later a slight decrease (48%) in infection coincided with a sharp decrease of Asterionella (35 cells/ml). On 5th February the decrease was reversed when infection declined sharply down to 8% and Asterionella to 23 cells/ml. After a further fortnight the infection was virtually absent (0.88%) and Asterionella reached its vernal maximum (2453 cells/ml). In early March infection rate increased to 17% but the numbers of the alga were almost unchanged (2391 cells/ml). After a fortnight a further increase in the infection (28%) and a sharp decrease in the numbers of Asterionella (941 cells/ml) were recorded. On the three subsequent fortnightly observations the low infection rates coincided with the further sharp decreases of Asterionella (down to 69 cells/ml). On 14th May the situation was most interesting

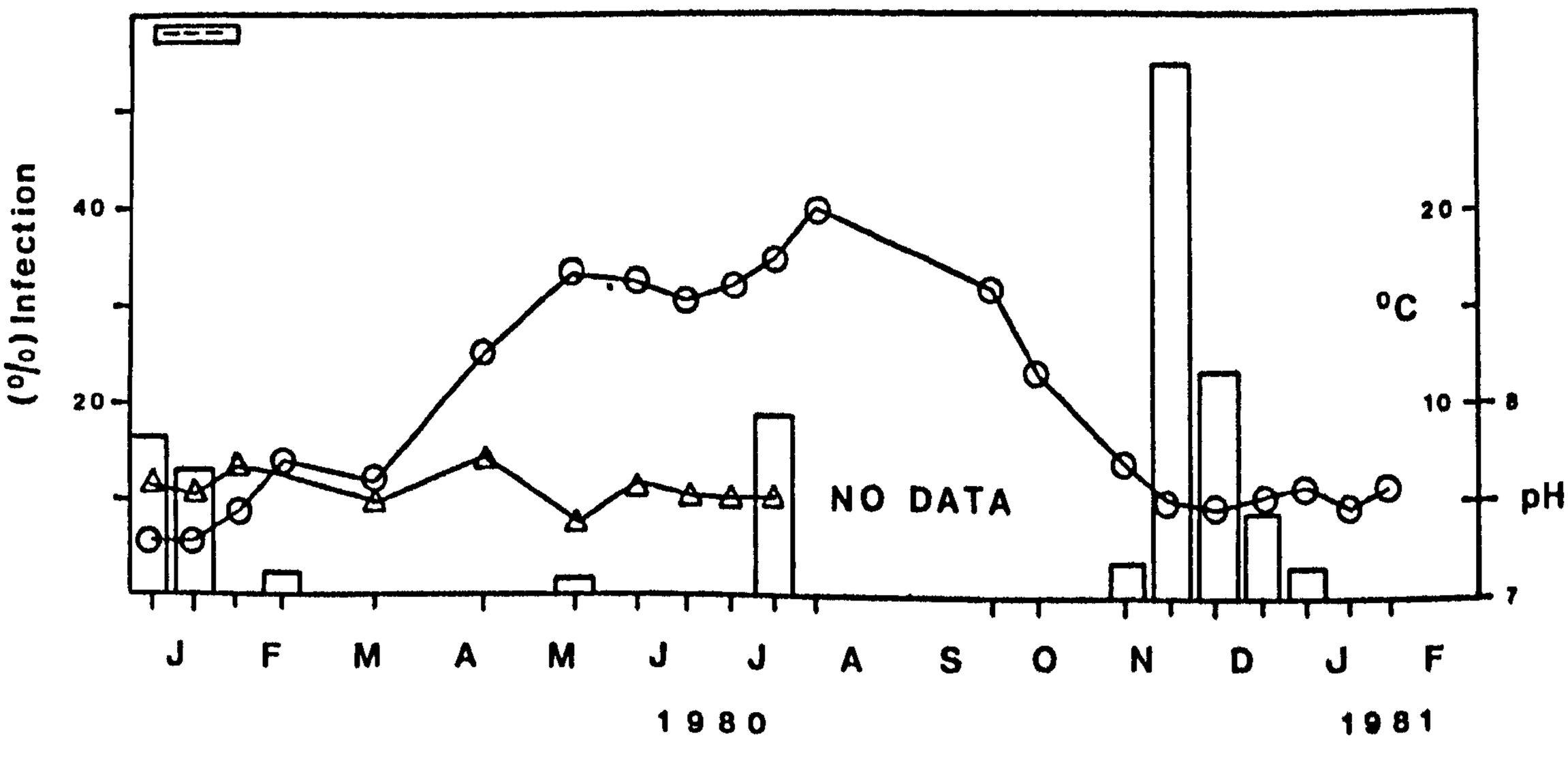
Fig.32. Epidemics of *Z. affluens* Canter on *A. formosa* in relation to physical factors.

□ % fungal infection

△——△ pH

○——○ temperature


(%)Infection
40
20
20
10
8
7
°C
pH
A M J J A S O N D
1978
(%)Infection
60
40
20
20
10
8
7
°C
pH
J F M A M J J A S O N D
1979

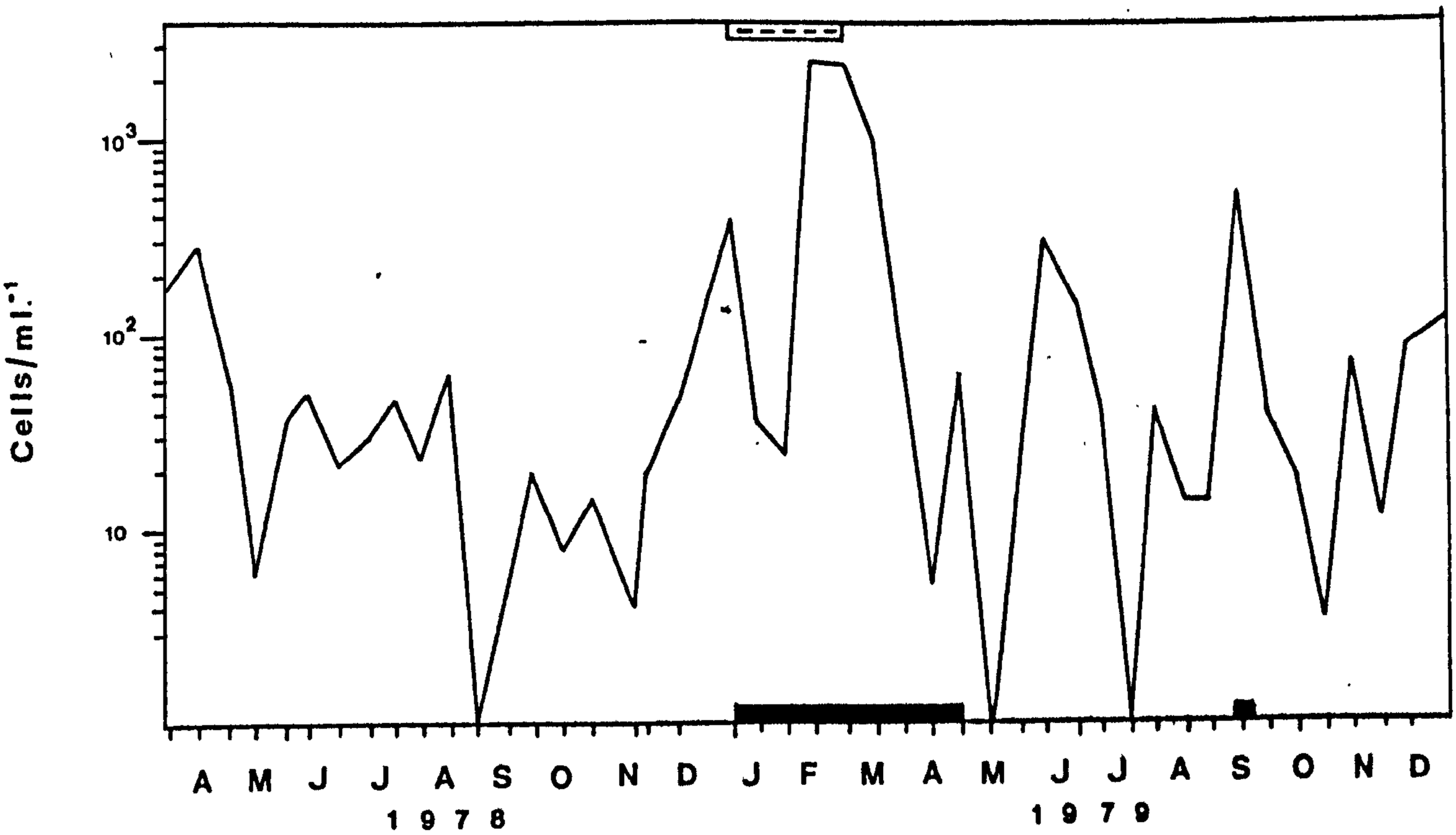

(%) Infection
40
20
20
10
8
7
°C
pH
NO DATA
J F M A M J J A S O N D J F
1980
1981

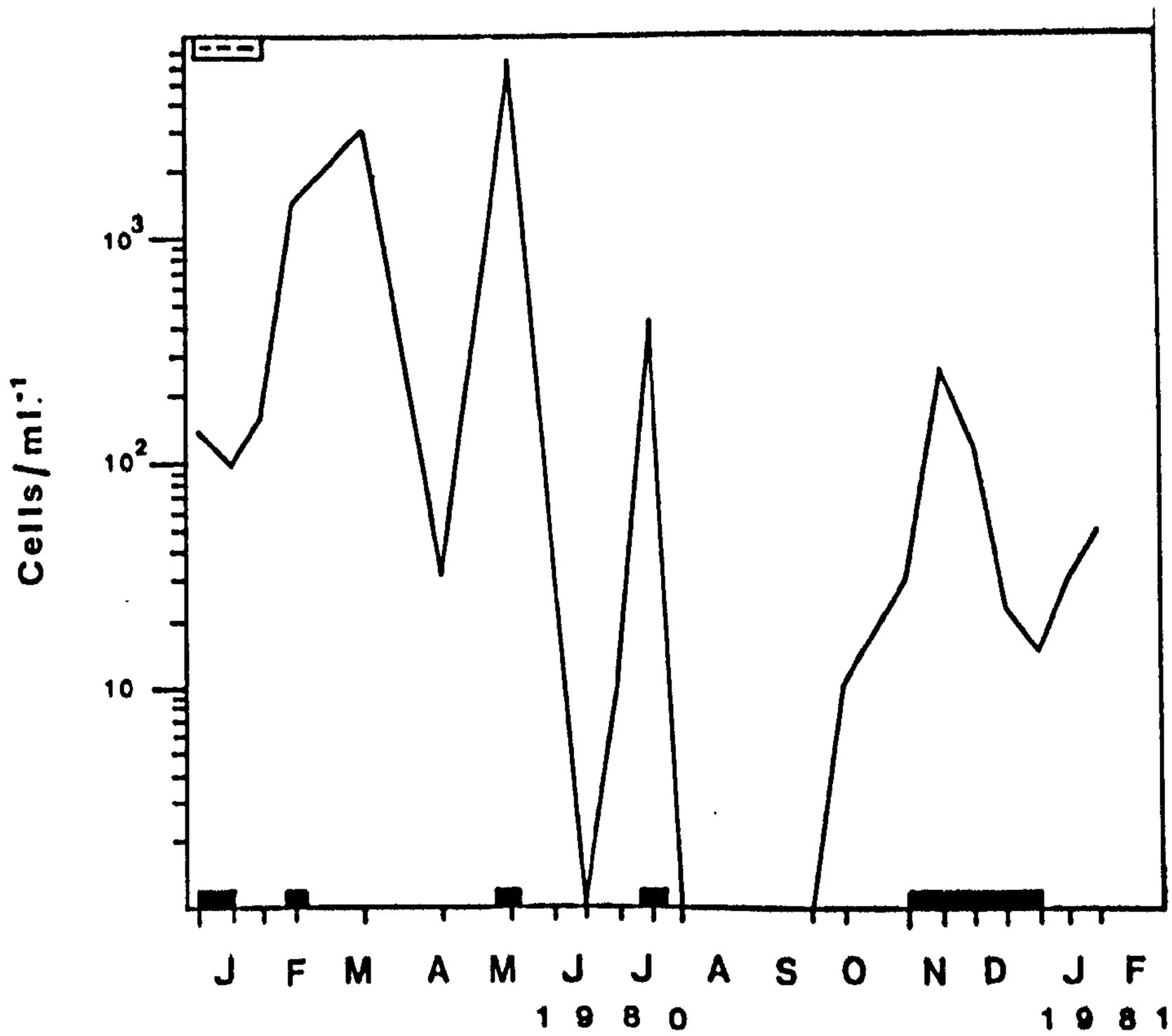
in that both the chytrid and Asterionella disappeared together. In the following months of 1979 the chytrid was absent although very low infections (0.92% and 0.88%) were recorded in September and in December respectively. Asterionella was present in the samples throughout this period.

In 1980, occurrence of the epidemic was recorded in January again. Winter duration of the chytrid was shorter and the infection rate was lower compared with the previous year. On 7th January 16% of the Asterionella cells were infected. After a fortnight the infection decreased slightly to 13% and a slight decline of the alga followed this. On the next observation, in the absence of the chytrid, the alga increased (160 cells/ml). On 18th February a few cells were infected (1%) and the response of Asterionella was not affected since an increase in cell numbers occurred (1486 cells/ml). On the next recording although there was no chytrid, numbers of the alga decreased dramatically down to 33 cells/ml due to an unknown cause. In May Asterionella reached its maximum (6548 cells/ml) - which was almost triple the size of the previous year - a few cells only were infected (1%). The chytrid was then absent until the middle of July. On 21st July the chytrid appeared on Asterionella with a slightly higher degree of infection (18%) than that of the winter epidemic. Asterionella at this time was present in a density of 446 cells/ml. After a fortnight both chytrid and alga disappeared and the chytrid was absent until the beginning of November. On 10th November only 3% of cells were infected. After a fortnight the maximum of the autumn epidemic was recorded

Fig.33. Fluctuation in the numbers of Asterionella formosa in relation to epidemics of Zygorhizidium affluens.

▬▬ periods of fungal epidemics


Cells/ml.-1
10^3
10^2
10
A M J J A S O N D J F M A M J J A S O N D
1978
1979


Cells/ml.-1
10^3
10^2
10
J F M A M J J A S O N D J F
1980
1981

(55%). Asterionella was then present at the rate of 272 cells/ml. After another fortnight the infection declined to 23% and Asterionella to 124 cells/ml. On 22nd December infection was down to 8% and Asterionella to 24 cells/ml. After a fortnight infection was only 3% and disappeared completely by the time of the next sampling when Asterionella started increasing towards its vernal maximum.

Comparison of Shearwater with English Lake District

In Shearwater, the occurrence of the epidemic of Z. affluens on Asterionella displayed a different pattern to the infections recorded by CANTER & LUND (1948, 1951, 1953).

Spring epidemics of the chytrid are usually characteristic of the English Lake District whereas only one extreme case was recorded in Shearwater within three years (Fig. 32). CANTER & LUND (1948) found that Asterionella and its parasite multiplied more vigorously in spring, but without any rise in the actual percentage of infected cells. The spring epidemic in Shearwater was much smaller compared with the winter and the autumn epidemics. In the English Lake District duration of spring epidemics were shorter than autumn ones but on most occasions the degree of the infection was greater (in some instances double the size of the autumn epidemic). The maximum of the spring epidemic in the present study was much smaller than those recorded in the English Lake District. In May (1980), however, very few Asterionella cells were parasitized by a sudden appearance of the chytrid but it disappeared with

equal rapidity.

Autumn epidemic also occurred only once in Shearwater (Fig. 32) although they occur more regularly in the English Lake District. There was no chytrid problem during the autumn of 1978. In 1979, two very slight infections were recorded in September and in December. However, an autumn epidemic was recorded in 1980, lasting for 2.5 months. 1980 was unusual in that two epidemics occurred and the autumn epidemic was much greater than that of winter (Fig.32).

Winter epidemics occurred in 1979 and in 1980 by the beginning of January (Fig.32). The former epidemic was much greater, in fact it was the greatest of all epidemics. Maxima of winter epidemics were recorded always in early January and disappeared by the end of February. Winter epidemic appeared to be more regular than other epidemics in Shearwater.

The summer period was not favourable for this fungus in Shearwater, In the first two years, the chytrid was completely absent from the samples during the summer (Fig.32). However, in 1980, a sudden appearance of the chytrid was recorded in the middle of July when 18% of cells were infected. Duration of the chytrid attack was very short, in fact in the next sample there was no sign of the chytrid on the alga at all. CANTER & LUND (1951) also reported a summer epidemic, occurring in June at a time when in most instances vernal maxima of Asterionella end. The summer epidemic in the English Lake District lasting only for a month but with a higher maximum of infection (69%) was greater than most spring and autumn

epidemics recorded in Shearwater.

Occurrence of Epidemics

The evanescent nature of the occurrence of chytrids was well documented by early works (COHN, 1853; ZOPH, 1888; DANGEARD, 1889b; WAGER, 1913; REYNOLDS, 1940 and others). The sudden appearance of Z. affluens on Asterionella formosa with high infection rates was also very characteristic in Shearwater. In the present study epidemics occurred when the host population was always increasing (generally during the vernal development of the host). In one case, however, the epidemic occurred while Asterionella was declining after vernal maximum. This supported CANTER & LUND (1948) who found that the occurrence of an epidemic occasionally may coincide, more or less closely with the period when the population is about to decline. CANTER (1954) also suggested that it is possible to forecast approximately when to find a particular fungal species in any one lake. CANTER found several of the common fungi (Amphicypellus elegans, Zygorhizidium melosirae) occurring at almost the same time of year. Constantly, in the present study, however, the periods of fungal epidemic was quite irregular and unpredictable - apart from two winter epidemics which occurred almost in the same period - over three studied years. They (1948) also suggested that the parasite is almost always present when the host is. The frequency of the parasite is too slight to reduce appreciably the numbers of Asterionella

for a large part of the year. In the present study although Asterionella was present during a large part of the year, the chytrid did not follow the cycle of Asterionella at all and apparently the parasite was absent for a long period of the year.

Occurrence of chytrid in relation to physical-chemical factors

A. Temperature; Correlations of temperature with the host and with chemical factors indicate that temperature is an indirect factor with respect to degree of parasitism. Temperature influences both biological and chemical factors. Concerning a direct relationship, although the fungus seems to be tolerant to a wide range of temperature, the main epidemics of Z. affluens coincided with very low temperatures (Fig. 32) in Shearwater.

In 1979, zoospores and sporangia were present under ice. In fact the overall highest infection level was recorded under ice. Apart from the summer epidemic, the maxima of other epidemics (main epidemics) occurred when the temperature was around 5°C or lower. Summer epidemics were exceptional in that occurrence of epidemics was at 17.5°C. However the encysted zoospores did not develop into a high percentage of sporangia. Infection consisted mainly of zoospores (Fig. 31). This is quite in contrast to other epidemics when the percentage of sporangia during the main epidemics was quite high (Fig. 31). In addition, the duration of the summer epidemic was the shortest (Fig. 32).

CANTER & LUND (1948) reported the wide temperature toleration for this fungus and also for R. planktonicum. PATERSON (1960) supported their view from his study of Rhizosiphon anabaena parasitic on Anabaena planktonica. They all agreed suggesting that temperature appears to have little direct effect on the chytrid maxima. In the present study, however, low temperature seems to be more favourable for the development of Zygorhizidium affluens than high temperature.

B. pH: pH level varied within a range of 7.28 - 7.59 during four epidemics of Z. affluens in Shearwater (Fig. 32). It is apparent from the same Fig. 32 that maxima of three epidemics occurred within a range of 7.51 - 7.52. However the winter epidemic in 1980 coincided with a slightly higher pH (7.59). Thus the present data would suggest that pH range of 7.52 - 7.59 was the most suitable for the development of this fungus. In addition, concerning the occurrence of maxima of three epidemics at 7.51 - 7.52 one might suggest that these values might be the optimum pH values for the parasite during its infection. PATERSON (1960) also reported the existence of an optimum pH (8.59 - 8.67) for the parasite Rhizosiphon anabaena during its infection on Anabaena planktonica.

C. Lake level: The occurrence of Z. affluens epidemics was always synchronous with increasing or high lake levels in the present study (Fig. 2). The percentage increase of infection also showed a distinct correlation with rising water level during the two most severe epidemics (Figs 2, 32) .However

further detailed data is required collected over many seasons before the occurrence and size of the epidemic can be definitely related to the changes in water chemistry etc. which are also involved when water level rises.

D. Turbulence: Turbulence of water will certainly have an effect on both host and its parasite. This effect will depend on the scale of turbulent water movements which is uncertain. However it is worth considering that even slight turbulence of quiet waters will have an effect on the free swimming zoospores since they are only about 3 - 4μ in length. Turbulence is a factor which has been studied only slightly but it has been shown to affect the growth of dinoflagellates in freshwaters (Pollingher, 19).

E. Light: The effect of the light intensity for the onset and size of epidemics is not clear either. There is no detailed data of the light intensity for Shearwater. However it is still possible to suggest, concerning the periods of epidemics that light has a slight effect on the occurrence of this fungus. Epidemics occurred within periods of increasing or long and high illuminations but these will be major factors affecting the hosts.

F. Dissolved Substances: Data of dissolved substances are available for only three epidemics.

Concentration of nitrate was always rising or high when all epidemics occurred (Figs 3,32). Silica and phosphorus

definitely displayed a different pattern than that of nitrate during epidemics (Figs 3 & 32) Occurrence of epidemics showed a correlation with sharp phosphate decreases or low phosphate concentrations. (Figs 3 & 32) The rate of the decrease in phosphate concentration was very similar during the two winter epidemics. Silica also showed a definite pattern in relation to epidemics in Shearwater. By the beginning of the first winter epidemic the concentration of silica was rising sharply but during the epidemic it decreased in an equal way. In 1980, silica displayed a similar pattern during winter epidemics but concentration was higher and decrease was slower (Figs 3 & 32).

In conclusion existence of a relationship between the occurrence of epidemics and the dissolved substances would be suggested by the present study since three basic nutrients displayed the same pattern for three epidemics. However, it is extremely difficult to separate out the effects of nutrients on the host and parasite and perhaps these nutrients mainly affect the rates of growth of the hosts.

G. Abundance of Asterionella: The importance of host availability and amount of host for the chytrid occurrence is now well known (previously explained).

In Shearwater Asterionella was present for a large part of the year over the study period, but epidemics occurred only in certain periods of the year. In addition, the numbers of the alga were always quite high when epidemics occurred (Fig. 33). It seems certain that high numbers of Asterionella provide a

greater chance for the zoospores to meet the host. However, this study would suggest that chytrids do not follow the cycle of Asterionella and the occurrence of epidemics requires high numbers of Asterionella to begin with.

This finding is a disagreement with the study of CANTER & LUND (1948) who stated that the parasite is always present when Asterionella is and LUND (1957) emphasized that the minimum Asterionella density for epidemics is about 10 cells/ml. The minimum Asterionella density was 86 cells/ml for epidemics in the present study. CANTER & LUND (1953) also suggested that density of Asterionella populations do not appear to affect the chances of an epidemic occurring since more epidemics were recorded at relatively low than at relatively high densities of Asterionella.

Importance of length of frustules

In the present study, the lengths of frustules of Asterionella showed a relationship with the fungal parasitism. The frustule lengths of cells of Asterionella ranged between 42 - 92μ. A composite table of the size distribution data suggests the presence of 4 - 5 size classes (Table 5) for Shearwater. As the numbers of some frustule size classes were not sufficiently high, only two distinct Asterionella populations are considered in this section. These size populations are named as Micro and Macro (Table 6). The micro population is composed of cells of 42 - 65μ length and the macro population of cells 68 - 92μ.

	Total no.of cells counted	42	44	47	49	52	55	57	60	63	65	68	71	73	76	78	81	84	86	89	92
11 July-12 Dec. 1978	333	-	-	30	69	92	73	61	8	-	-	-	-	-	-	-	-	-	-	-	-
Jan - Dec. 1979	1627	-	12	27	129	520	531	171	24	4	12	23	5	4	6	30	10	21	40	42	16
Jan - Dec. 1980	1147	2	-	38	142	283	183	30	26	-	-	8	-	10	55	189	136	40	5	-	-

Table 5. Distribution of cells in accordance with frustule lengths.

Size of class	Micro population	Macro population
Mean length of cell	53µ	80µ
Range in length	42 - 65µ	68 - 92µ

Table 6. Size classification of Asterionella formosa cells based on frustule length

The Micro population was always present while the Macro population was recorded only during certain Asterionella periods (Fig.34). Population dominance varied in different periods of the year but generally the micro population exceeded the macro population and its occurrence was more constant.

During the two fungal epidemics the macro population was present alongside the micro population (Figs 34,35). During the first epidemic (1979) the macro population was very low in numbers compared with the micro population, and no infection was recorded at all. January 1980 provided a good example to compare the infection level of both populations. During this period the numbers of both micro and macro populations were quite high. Although both populations were bearing the chytrid thalli, infection level on the micro population was much greater than on the macro population. Moreover, at one stage the macro population was greater than the micro population in numbers but still the infection level was very low on them compared with the infection of the micro population.

It is very clear from the Fig.36 that infections of the three major frustule lengths - 49µ, 52µ, 55µ - of the micro population were the most considerable out of all infected frustule lengths. (See Table 5). Numbers of other frustule lengths in the micro population were comparatively very low. Therefore one should consider the abundance of these frustule lengths for their high infection rates in the samples (See Table 5). Obviously the chytrid would have more chance to meet these three dominant frustules than others. One should

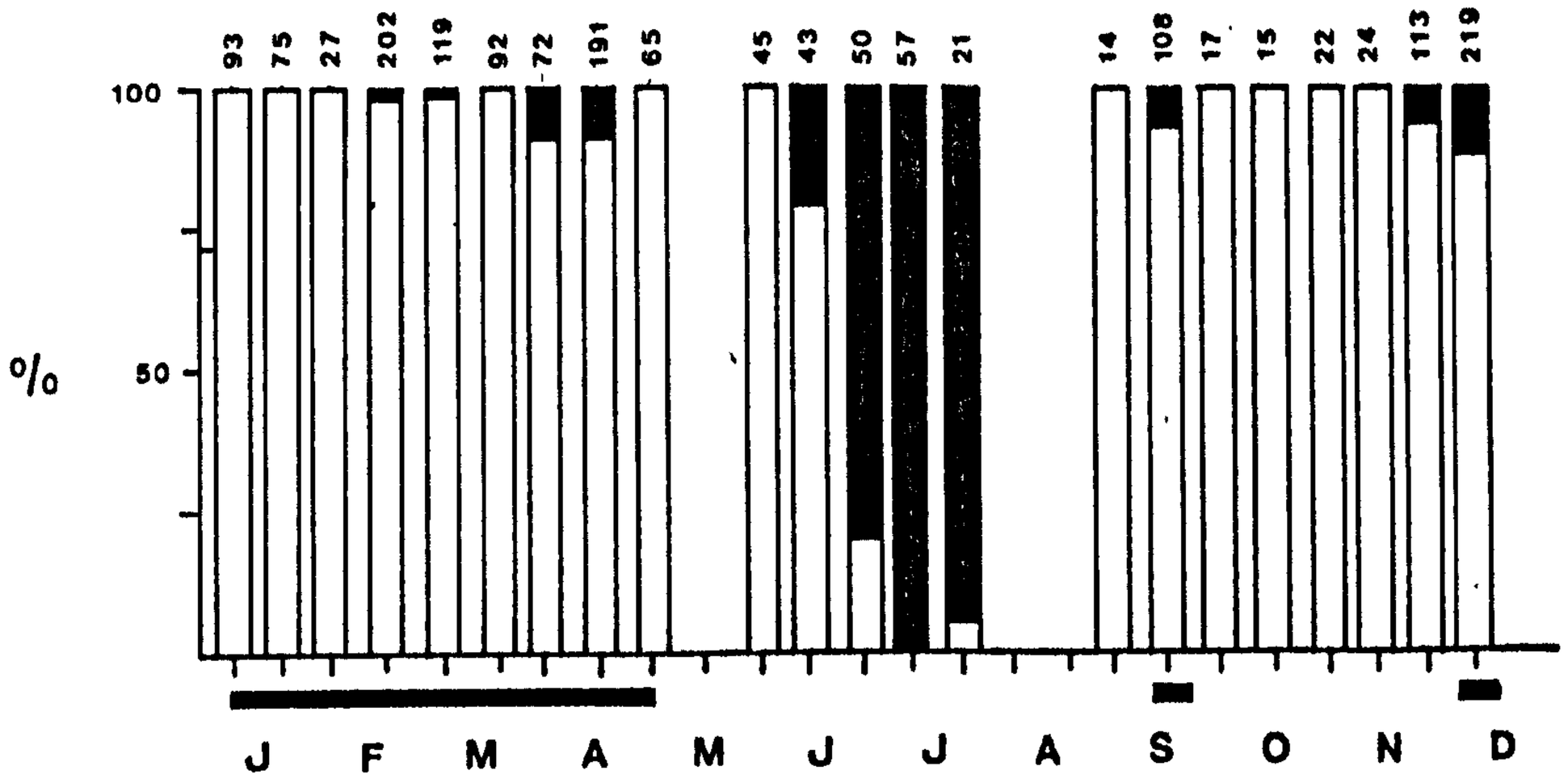
Fig.34. Seasonal distribution of Micro and Macro population of A. formosa.

□ Micro population

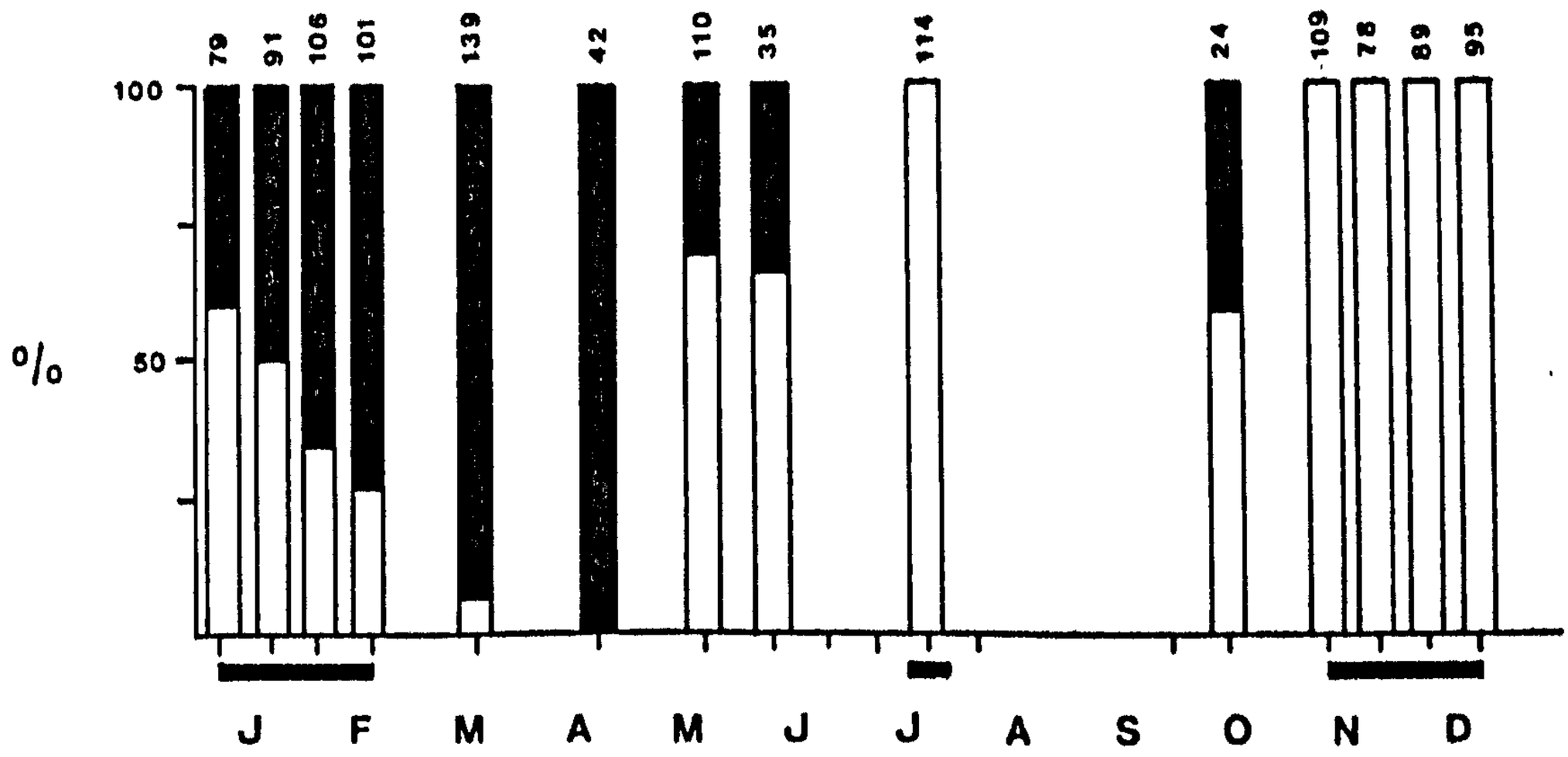
■ Macro population

▬▬ (periods of fungal infection)

Note: Numbers indicate the total number of cells counted.


93
75
27
202
119
92
72
191
65
45
43
50
57
21
14
108
17
15
22
24
113
219
100
50
%
J
F
M
A
M
J
J
A
S
O
N
D

1979


79
91
106
101
139
42
110
35
114
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78
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%
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S
O
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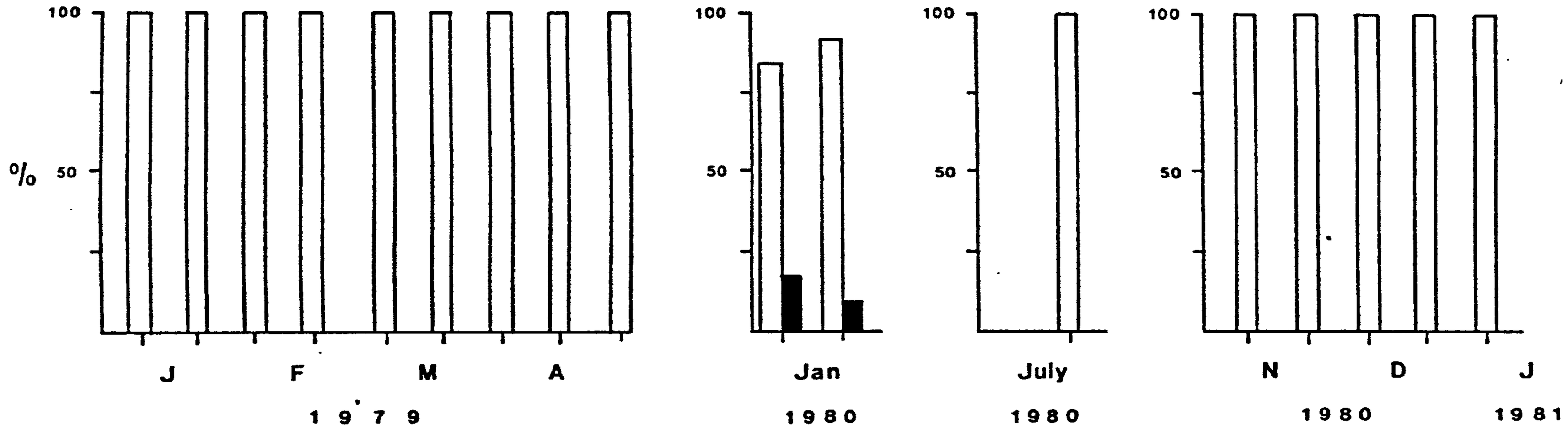
D

1980

Fig.35. A. % infection of micro (□) and macro (■) populations of A. formosa during epidemics.

B. Total distribution of micro (□) and macro (■) populations during epidemics.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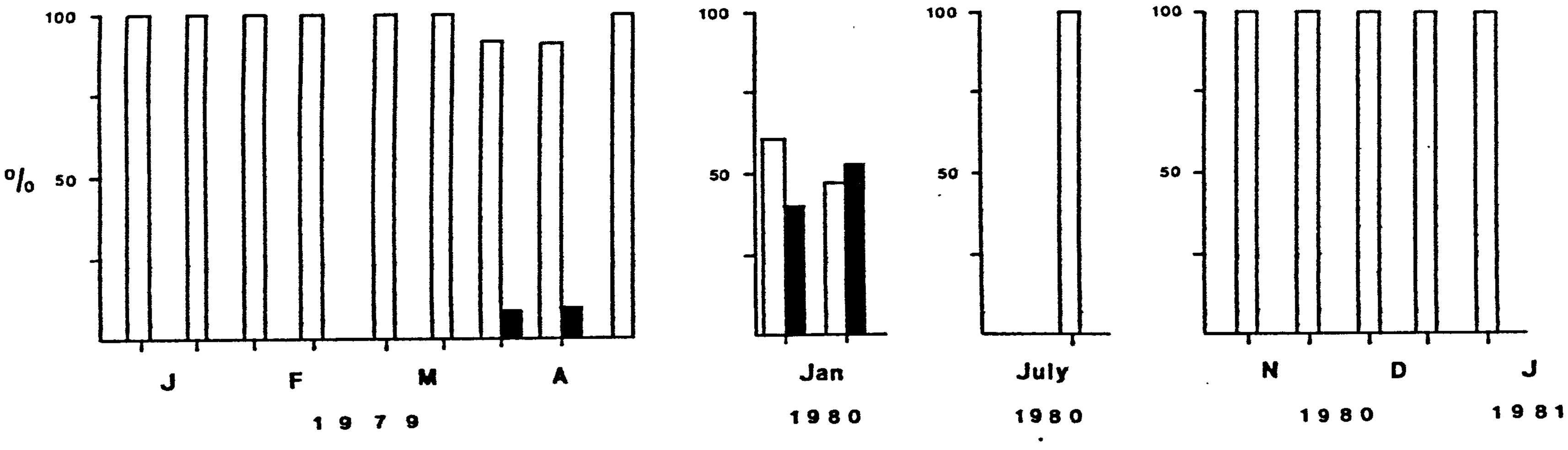
1981

A


100
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1980
1981

B

also remember that these frustules (49μ, 52μ, 55μ) were still attacked more heavily than others during the period when the numbers of these frustules were much lower than the numbers of other frustules. Infections on the smallest and the largest frustules were very low indeed.

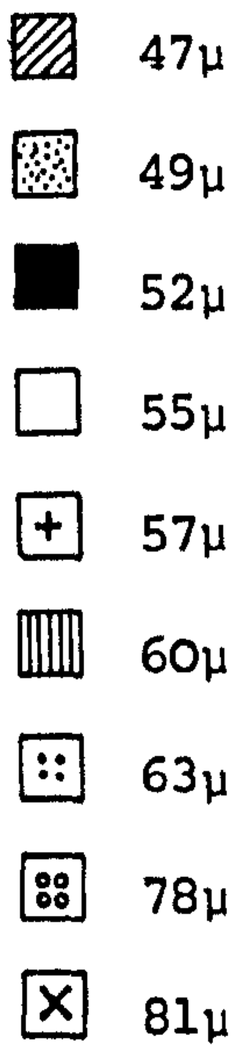
In conclusion, the present data suggests that a distinct frustule size of Asterionella from 49μ to 57μ in the case of this lake study is more susceptible to fungus attack than smaller and larger frustules whatever the host population size.

KOOB (1966) also documented the occurrence of five distinct populations of Asterionella - based on frustule length - for two lakes in Colorado. The lengths of the frustules ranged from 43μ to 103μ. The β population (50 - 67 μ) was important in both spring and fall blooms and differed from the other population in that it succumbed to attack by Rhizophydium planktonicum Canter, while the rest remained fungus-free.

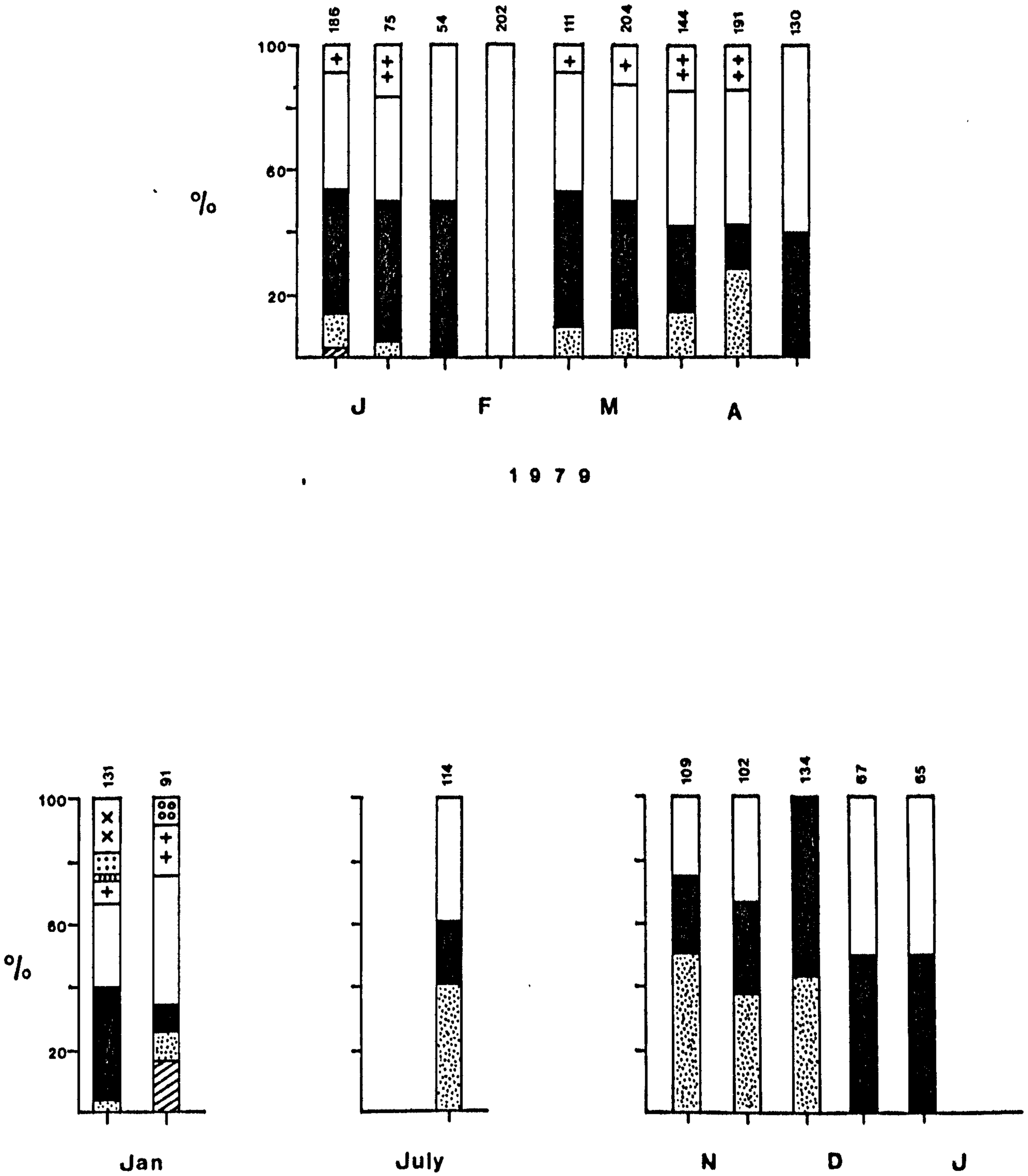
KOOB's study was interesting in that as in the case of Shearwater, a distinct frustule size was susceptible to fungus attack, while smaller and larger cells were not. Eventually he suggested that there was no marked effect of parasitism on the algal population since the host increased or decreased in numbers whether the parasite was present or not.

It is clear that the present study is in accord with KOOB's study in that the frustule length of Asterionella is important for the fungal parasite but also in disagreement since the parasite does reduce the rate of population growth.

Fig.36. Distribution of the length of the infected frustules of A. formosa during epidemics of Z. affluens.



Numbers indicate the total number of cells counted.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75
54
202
111
204
144
191
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100
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20
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102
134
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Jan
July
N
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1980

Absence of Z. affluens

Factors which are responsible for the absence of Z. affluens during a large part of the year are unknown. Moreover, without determining the factors controlling the onset of epidemics, it is quite difficult to make any suggestions about which factors are responsible for the absence of the fungus. However, the absence of Z. affluens always coincided with high temperatures in Shearwater (Fig. 32). CANTER & LUND (1948) attributed the absence of epidemics to the relatively faster growth of Asterionella during the spring growth period. However the present study also showed that the spring epidemic might occur regardless of the growth rate of Asterionella.

Nothing is known about organisms which might feed on the zoospores or attached stages of the fungus. During all epidemics, however, zooplankters were virtually absent in Shearwater. A few Bicosoeca and Salpingoeca were present on Asterionella cells during some epidemics. However these organisms feed on bacteria, but could possibly ingest fungal zoospores.

In conclusion, no single factor or groups of factors seems to control the onset of an epidemic nor the absence of the fungus. This conclusion is in harmony with CANTER & LUND (1948).

Effects of Parasitism on Asterionella Population

A Epidemics of Z. affluens affecting the growth of Asterionella are given in Fig. 33 . It is obvious from this figure that the vernal development of this alga was interrupted during the two most severe epidemics (see Fig. 32 for epidemics). Before the onset of these two epidemics (winter 1979 and autumn 1980) the development of Asterionella was very rapid for the vernal maximum. The occurrence of epidemics with an increasing number of cells attached to the host resulted in a rapid decline in cell numbers (383 - 37 cells/ml, 1979; 272 to 24 cells/ml, 1980) which continued throughout the epidemics. Numbers of the alga then started increasing again. In July 1980, another sharp decrease in the numbers of the alga was recorded after a less severe fungal epidemic. Asterionella reached its highest cell number for thistime of year in any of the four years. Although there was always a decline in the numbers of the alga in July due to low or virtual absence of silica, fungal parasitism appeared to force the alga to decline. The decline was the sharpest of all four years during this summer epidemic. In fact, 1978 provided a perfect chance to compare the influence of parasitism on the alga. In this year during the vernal development of Asterionella the chytrid was absent. Onset of the vernal maximum started as usual as early as November and reached a maximum after a continuous increase. There was no interruption or decrease during the vernal development at all. Silica concentration appears to have a role in this situation since it was overall the highest of all years.

However, silica was always rich enough to support the growth of Asterionella during the vernal development but cell numbers of Asterionella decreased sharply under the effect of parasitism.

However the Asterionella population did not always decline during the periods of epidemics in Shearwater (Fig. 32). During the spring epidemic (1979) the alga reached its maximum in spite of the presence of fungal parasites and the subsequent decrease of the alga during the same epidemic seemed to be due to exhausted silica concentration. Even so, there was an effect of the parasite masked by the effect of the silica.

In addition, during the winter epidemic of 1980 (Fig.33) there was no serious interruption from parasitism although 16 % of cells were infected. There was only a slight decrease after the maximum of the epidemic which lasted two months.

In conclusion, present data supports the view of CANTER & LUND (1948) that at certain times the fungi are capable of reducing the number of Asterionella cells. It is certain from Figs.32,33 that the restriction on the rate of increase of Asterionella appears to be related to the degree of infection of the population rather than to the duration of epidemics. Sharpest declines in the numbers of Asterionella always coincided with very severe epidemics whereas when the degree of infection was under 20% it was not really a threat to the Asterionella. During such periods silica concentration was always sufficient for the growth of the alga. In addition the numbers of Asterionella always increased after the end of an epidemic showing that epidemics exert only a mild detrimental effect on the alga.

CANTER & LUND (1948) suggested that other factors obviously might be reducing the algal numbers at the same time as the fungus. They usually found concentrations of nitrate, silica and phosphate high or rising during periods of epidemic parasitism on Asterionella. In addition they also noted that parasitism of Asterionella tended to be slight when inorganic matter was low. A slight correlation has been found in the present study between the decrease of numbers of Asterionella and the chemical data during fungal epidemics. In periods of algal decline during severe parasitism by Z. affluens, silica concentration was always favouring the alga. Silica was either rising sharply or already high. Thus silica is most unlikely to be the cause of decrease in the Asterionella population. Nevertheless, rapidly declining or low phosphate concentration tended to coincide with the decrease of the numbers of Asterionella during such periods. Therefore phosphate seems to be the only chemical property which might be involved in the decrease. In the case of nitrogen, it was always sufficient to support the development of Asterionella. HUTCHINSON (1944) discussed the causes of decline of phytoplankton maxima giving evidence that a depletion of mineral substances is one of the major causes for such decreases in algal population. This view, however, explains the declination of phytoplankton during a normal cycle but it cannot apply to Asterionella during epidemics since all the basic nutrients were present in sufficient quantity to support the growth of Asterionella.

Physical factors seem to be combining to induce the decline of Asterionella during epidemics.

Temperature

Dependency of Asterionella on temperature within certain limits was shown by LUND (1949). In cultures Asterionella grew very slowly or declined when the temperature was low or lowered. However LUND (1950) also reported that Asterionella can grow actively within the range 1.5 - 24°C in British lakes. Temperature displayed almost a regular cycle over the study period of Shearwater. However Asterionella was quite tolerant of low temperatures in Shearwater.

Onset of the vernal maxima of this alga coincided with a temperature decrease. Ice formation in Shearwater may not account for the decrease of the algal population in 1979 during severe epidemic. In 1978, during the same period, Shearwater was also covered with ice (with longer duration) but there was no decrease in the numbers of Asterionella at all.

The suggestion of LUND (1963) that the wetter and the colder the winter, the smaller size of Asterionella maximum appears to be untenable for Shearwater. Three successive winters were quite cold but the size of maxima varied very much. Thus the smallest vernal maximum in 1979 may not really be attributed to cold weather but may be due to parasitism and lack of silica. Consequently the temperature seems to be unlikely to be responsible for the decrease in the numbers of alga.

pH.

The hydrogen - ion concentration in the aquatic environment may act on phytoplankton as a controlling factor according

to its level. However, variations in pH level were very small in Shearwater and unlikely to affect the development of Asterionella at any time (Fig. 1).

Light and turbulence

Light and turbulence may also limit the number of cells of Asterionella (CANTER & LUND, 1948). The importance of light was confirmed by LUND (1949) and shown to influence the rate of development of Asterionella blooms. No comment can be made on this point since there is no available light data for Shearwater. In the case of turbulence similarity of autumn and winter periods may not allow any suggestions to be made.

Lake level

Occurrence of fungal epidemics synchronized with increasing or high water levels in Shearwater. CANTER & LUND (1948, 1951) also found that during epidemics the lake level rose rapidly because of floods and thus cell numbers of Asterionella may also be reduced due to a mechanical depletion. This may not be the answer for the reduction of the alga during epidemics of Shearwater. In 1978 there was no epidemic at all. The water level was quite high - in fact the highest of all years - while Asterionella was increasing rapidly for the vernal maximum and there was no restriction on the numbers of Asterionella at all.

Competition

Competition between diatoms during vernal blooms was a factor since it wouldoperate via the overall nutrient supply. Only centric diatoms appear to be a serious rival for Asterionella. However in the periods when the main epidemics occurred during which numbers of Asterionella decreased, the concentration of

silica, nitrate and phosphate were generally sufficient to support the growth of diatoms.

Grazing

Grazing by zooplankters was not really important in Shearwater. Numbers of zooplankters were usually low or absent for a large period of the year. However, none of the zooplankters of Shearwater are reported to graze on Asterionella. Moreover, the zooplankters were virtually absent during epidemics.

Effects of parasitism on vernal maximum of Asterionella

B. CANTER & LUND (1951) do suggest that the parasites may delay the time of maximum algal number or may decrease the size of the maximum in the case of Asterionella.

In 1978, without the presence of the chytrid Asterionella reached its maximum on 20th March in Shearwater with a considerable population (3925 cells/ml). In the following year the infection of the chytrid was very severe but the vernal maximum of Asterionella occurred a month earlier (19th February) than that of 1978. However the vernal maximum was much smaller (2453 cells/ml). In 1980, during the vernal development of the alga, the degree of infection was much less and shorter than that of the previous year. Despite the infection Asterionella reached 3183 cells/ml on 17th March. This was thought to be the vernal maximum but the alga then declined to 33 cells/ml due to an unknown event and a second vernal maximum was recorded. This was also the latest occurring of all the spring maxima, on 20th May, and the largest (6548 cells/ml).

Thus the present study is in harmony with LUND & CANTER (1981) but the growths in Shearwater are less precise than those in the Lake District. Effects of parasitism on the vernal maximum of Asterionella are confusing in Shearwater as they are not compatible over the study period. First of all the smaller size of vernal maximum in 1979 could be also attributed to low silica concentration as well as to parasitism. Silica concentration was the lowest and the parasitism was the most severe compared with other years. Therefore the effect of parasitism on the vernal maximum was certainly marked by silica. Secondly, the sharp decrease and delay during the vernal development in 1980 tends to be due to unknown factors which may be chemical (phosphate) but it is unlikely that it is parasitism. During this period chytrids were absent.

In conclusion, the present data would suggest that the numbers and vernal maximum of Asterionella formosa are obviously affected by parasitism as well as other factors but the degree of the correlation of these factors remains obscure. Similar suggestions are also on record (CANTER & LUND, 1948, 1951; LUND, 1950) that changes in the density of populations reflect both the effects of parasitism and other factors. The small size of Shearwater may result in more rapid fluctuations in physico-chemical factors than would be recorded in the larger English Lake District lakes which are thus better buffered.

Variations of live cells per colony

C. Variation of live cells per colony in Asterionella formosa

is well known (GARDINER, 1940; PEARSALL, GARDINER & GREENSHILDS, 1946) particularly during periods of active growth. It falls as environmental conditions become less favourable. CANTER & LUND (1948) stated for the first time that the average number of live cells per Asterionella colony decreases in all epidemics. In addition, an increase was observed after the end of an epidemic in their study.

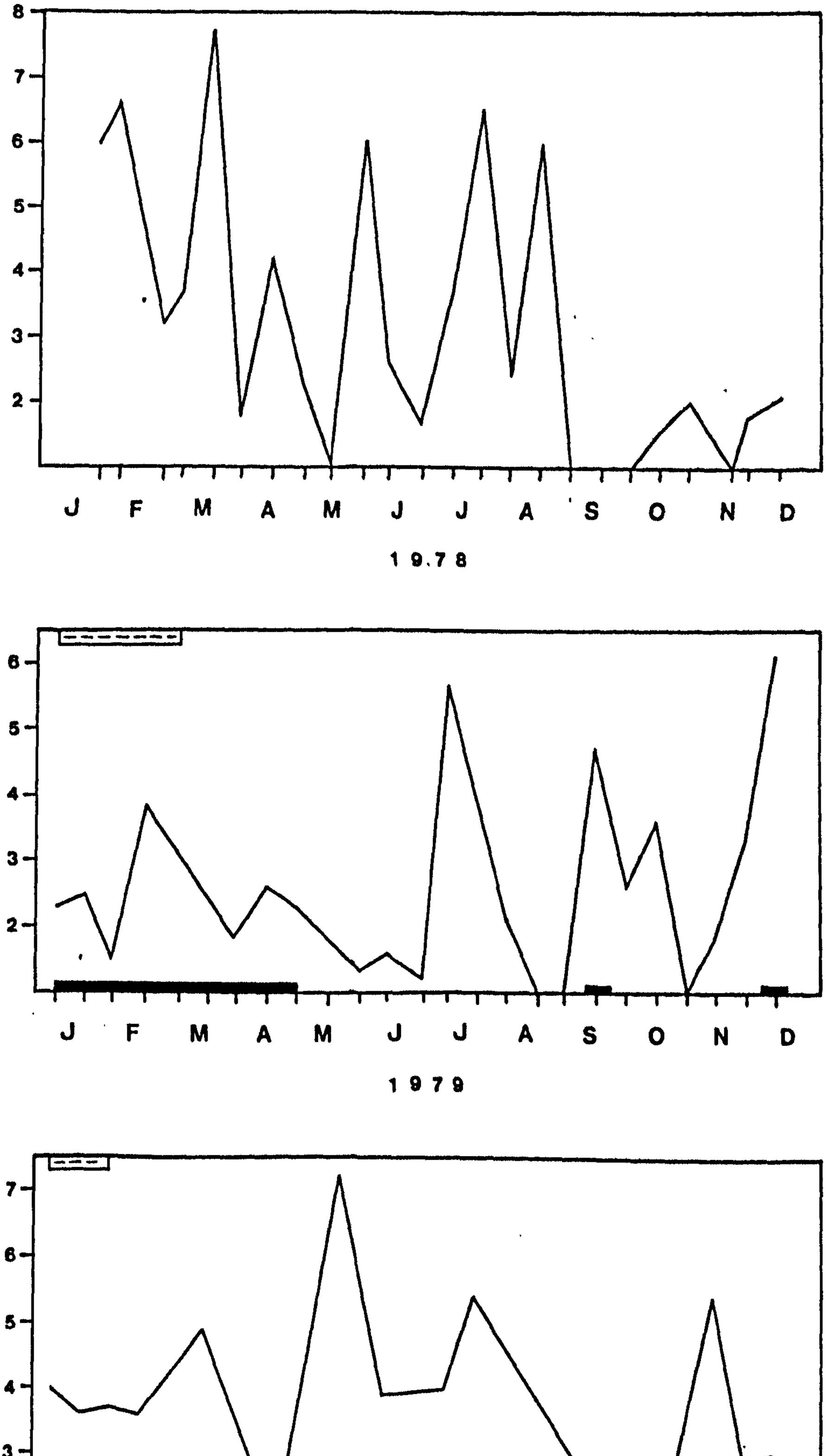
Fluctuations in the average number of cells per Asterionella colony during epidemics in Shearwater are shown in Fig. 37.

In 1979, before the epidemic started, average numbers of cells per colony was increasing gradually and continued to increase despite the high infection which was the most extreme recorded. However, towards the middle stage of the epidemic (low infection rate) a decrease was followed by an increase towards the end of the epidemic. Then a second epidemic started without an interval of recovery. During the period of high percentage infection, the average numbers of cells per colony decreased continuously. When the infection subsided a slight increase was recorded. It is noteworthy that the number of cells per colony continued to decrease after the epidemic ended which is quite in contrast to the finding of CANTER & LUND (1948)

In 1980, the average number of cells per colony was decreasing before the winter epidemic started. A slight increase took place during the epidemic and a sharper increase occurred after the epidemic. The summer epidemic was exceptional in that during this short epidemic cells per colony

Fig.37. Seasonal variations of the average number of cells per Asterionella colony.

(▬▬) periods of fungal infection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J F M A M J J A S O N D
1978
6
5
4
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2
J F M A M J J A S O N D
1979
7
6
5
4
3
2
J F M A M J J A S O N D
1980
No. cells per col.

continued to increase. No Asterionella cells were present in the sample after this epidemic. The last epidemic, however, showed a better correlation between the development of the epidemic and the cell number per colony. There was a gradual decline until the end of epidemic and then numbers rose again afterwards. This is exactly what was described by CANTER & LUND (1948), but it occurred only in one epidemic. Moreover, the average numbers of cells per colony was decreasing again after the epidemic. Thus none of the epidemics in Shearwater showed a similar pattern to those of CANTER & LUND (1948).

In conclusion findings in the present study concerning the average number of cells per colony are somewhat in disagreement with CANTER & LUND (1948). The average number of cells per Asterionella colony did not show a definite correlation with epidemics. Numbers decreased or increased - regardless of the severity of the epidemic - during epidemics of Z. affluens. In addition there was generally a decrease in the average number of cells per colony after epidemics ended. However irregular fluctuations of average number of cells per colony occurred over the study period (Fig. 37).

Effect of parasitism on the health of Asterionella cells

D. In all epidemics numbers of dead cells increased in Asterionella colonies. Fig. 38 clearly shows the severe effect of parasitism on healthy cells. Before epidemics began, numbers of dead cells were negligible in the samples. By the appearance

Fig.38. Distribution of healthy and dead cells of <u>Asterionella</u> during periods of epidemics.

A (□) healthy infected cells

(■) dead infected cells

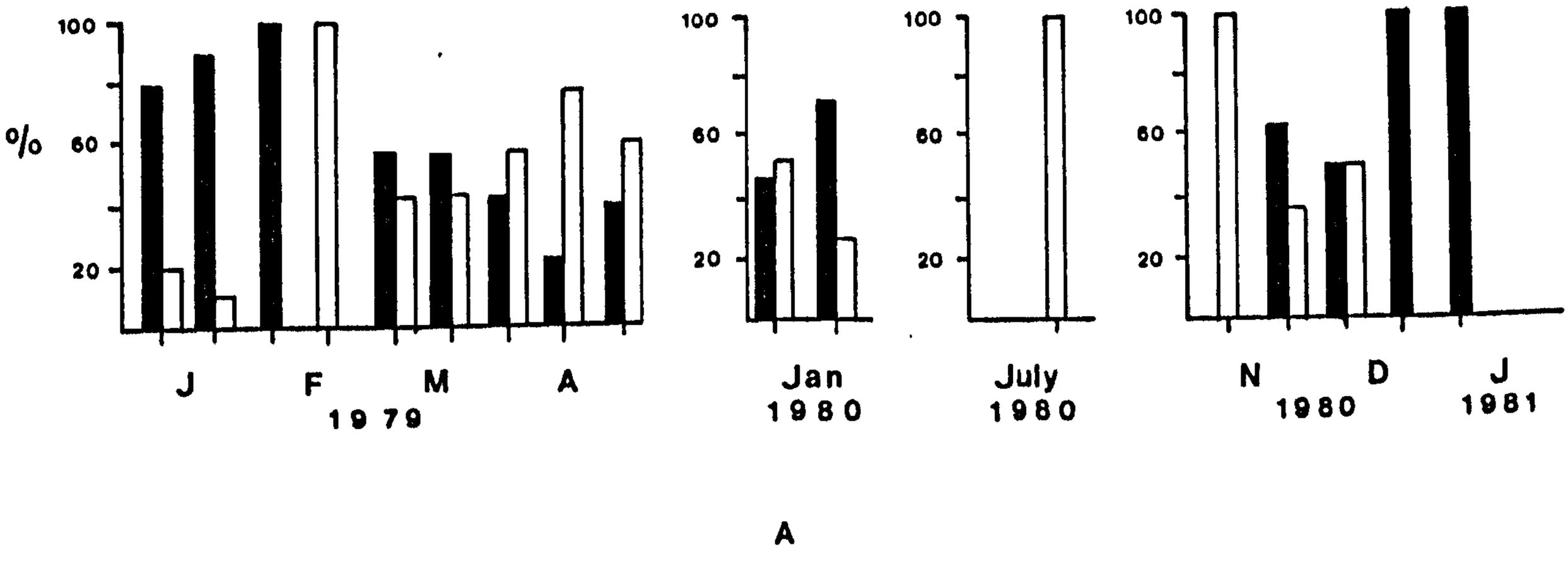
B (□) healthy uninfected cells

(■) dead uninfected cells

C (□) total (infected and uninfected) healthy cells

(■) total (infected and uninfected) dead cells

<u>Note:</u> TNC indicates the total number of cells counted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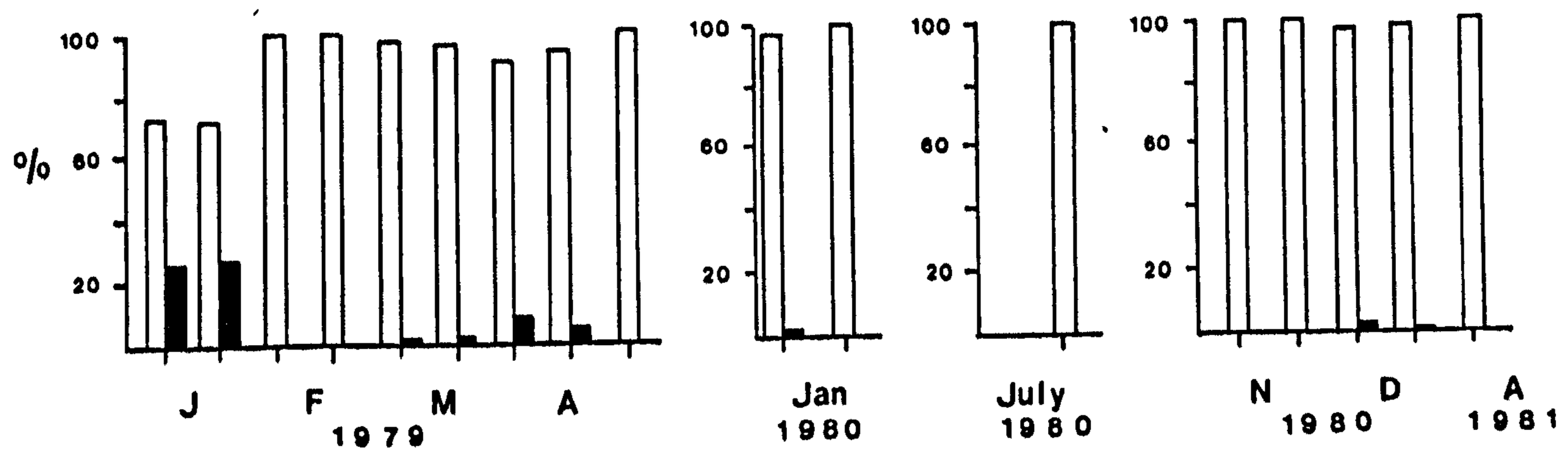
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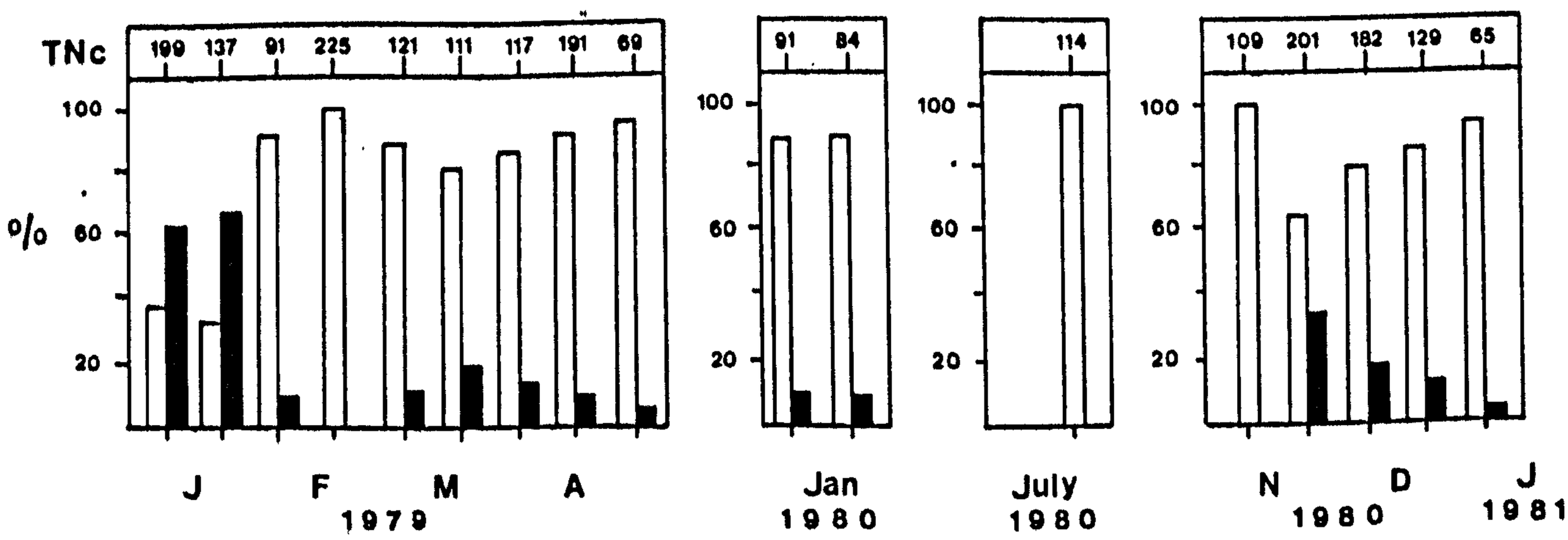
TNc
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of chytridson Asterionella cells, numbers of dead increased sharply due to infection. Parasitized cells were generally dead while the uninfected cells in the same colony remained healthy (Fig.30p). At one stage the effect was so drastic that numbers of dead cells outnumbered healthy cells. This was observed during the most severe epidemic of all when the number of dead cells was almost double that of healthy cells (Fig. 38).

Numbers of development stages of Z. affluens during all epidemics were analysed and are shown in Fig. 31. It is apparent from this Fig. 31 and Fig. 38 that cells bearing encysted zoospores were still healthy. This is because the "feeding" by encysted zoospores on the protoplasm of Asterionella cells had only just started and cells are still very little disorganized. A drastic effect was observed on the cells bearing mature or empty sporangia and resting spores (zygots) . As the zoospores became sporangia most of the Asterionella cells died increasing the ratio of dead cells (Figs 31,38).

Present results support the view of CANTER & LUND (1948, 1951) that during all epidemics of fungi the ratio of dead cells to live cells increases.

Advantages of parasitism - Parasitism favours the growth of other diatoms

E. The view of parasitism of one alga favouring the development of another (CANTER & LUND, 1951) is fully supported in the case of Asterionella in the present study. Decline of

Asterionella during severe epidemics favoured the growth of other diatoms in Shearwater. In fact, 1979 and 1980 provided good examples of competing diatoms.

In 1979, without the presence of parasitism, centric diatoms (Cyclotella, Stephanodiscus) failed to develop during the rapid growth of Asterionella. Despite the high silica, nitrate and phosphate, centric diatoms were increasing after this period.

However, in the following year during the vernal development an interesting situation was observed. Decline of Asterionella during the severe epidemic of Z. affluens appeared to favour the growth of centric diatoms. Numbers of centric diatoms had been declining but suddenly they started increasing rapidly - in contrast to the previous year. In fact, centric diatoms reached their highest recorded number for this period of the year. The size was almost equal to vernal maximum of Asterionella (Fig.13) although the silica was at its lowest level of all years. In this case parasitism of Asterionella seemed to enable the centric diatoms to increase before the supply of silica became limiting.

In 1980, in the period of the second most severe epidemic of Z. affluens (November - January), the numbers of centric diatoms were increasing very sharply in contrast to previous years while Asterionella was decreasing. As a matter of fact, this was the sharpest increase in the numbers of centric diatoms for this period of the year out of all years studied. although the numbers of centric diatoms were also high in previous years.

In addition there was an unusual peak of Melosira granulata (Fig.15) during the period of November - January 1980,

and the appearance of Nitzschia in the samples in the periods of epidemics might also be due to the decline of Asterionella.

Summary and conclusions

Occurrence of epidemics of Z. affluens on the Asterionella population was unpredictable. However, three major epidemics occurred during the vernal development of Asterionella.

Main epidemics lasted for two months on each occasion.

Chytrids could occur any time of the year thus making it very difficult to correlate the factors determining the occurrence of the chytrid.

Temperature seems to be the only stable factor affecting the occurrence of epidemics while the effects of the remaining physical-chemical factors is obscure. Occurrence of epidemics generally coincided with very cold periods and absence of chytrid with high temperatures.

High numbers of Asterionella might be an important factor for the occurrence of epidemics since they were always recorded when Asterionella numbers were high.

Parasitism is clearly one of the factors responsible for the decline in the numbers of Asterionella. In addition the decreasing growth rate might be attributed to the degree of infection.

It would seem to be logical to suggest that development of Asterionella would continue to increase greater than it did judging from its growth rate if parasitism was absent.

Z. affluens is certainly a parasite appearing on actively growing healthy Asterionella populations.

Z. affluens shows a definite host specifity since it always appeared only on Asterionella. Centric diatoms were

also in high numbers during such periods but were not infected by this chytrid.

The degree of infection depends on the relative growth rate of both host and parasite.

The epidemics do not eliminate the Asterionella populations and Asterionella increases after the most severe epidemics.

It might be possible to forecast when epidemics will occur since occurrence generally coincided with the period of vernal development of Asterionella.

Average number of Asterionella cells per colony did not correlate well with the development of an epidemic since either a decrease or an increase occurred. In addition average numbers did not increase after the epidemic.

The highest infection was 63.72% over the study period. In contrast, up to 90% infection was recorded by CANTER & LUND (1951).

Fungal Parasitism of Fragilaria crotonensis Kitton

Fragilaria crotonensis was an important phytoplankter of Shearwater and the filaments were encountered almost throughout the year (Fig. 7). It commonly reached its maximum during autumn (October - November) and summer (June) while in winter but not always during spring, it was usually present in small numbers relative to those of Asterionella (Figs 5 & 7). During this study, the filaments of Fragilaria succumbed to heavy fungal attacks at certain times of the year.

CANTER (1950) has described a new chytrid, Rhizophydium fragilariae Canter and assigned it to F. crotonensis as it has never been observed on any other planktonic diatom in the English Lake District. F. crotonensis is also infected by Chytridium versatile Scherffel in the English Lake District but R. fragilariae causes the major decreases in diatom numbers and occurs for much longer periods (CANTER & LUND, 1953). Another two species, species 3 and species 4, about which little is known at present, were reported by the same authors infecting the diatom from other parts of Great Britain and Europe. Massive infection by R. fragilariae on filaments of F. crotonensis was reported by PONGRATZ (1966) in Lac Léman, Switzerland. JOHNSON (1967) tentatively referred to R. fragilariae, sporangia which he found growing on anotherdiatom Rhizosolenia in saline habitats. Due to certain morphological differences, CANTER & JAWÒRSKI (1982) do not consider that this is the same taxon. R. fragilariae was also found on F. crotonensis and F. capucina Desmariéz growing

in lakes in the U.S.A. (PATERSON, 1958; 1967).

In the present study, identification of the fungus infecting F. crotonensis and its resemblence to R. fragilariae prompted further observations. The present fungus was mostly either at encysted zoospore or sporangial stages therefore its full life cycle could not be observed. Attempts to culture this fungus were unsuccessful.

Two morphologically distinct forms of F. crotonensis have recently been reported by CANTER & JAWORSKI (1982): 1. Rod-type (cells with narrow ends); 2. Flared-type (cells, flaring out at their extremities); from the phytoplankton of Windermere. In the same study it was quite interesting that rod-type forms were infected by R. fragilariae while flared-types were colonised by species 3, although the two forms existed together. However, occasional sporangia can occur in both cases, on cells of the opposite kind. In addition CANTER & JAWORSKI (1982) also stated that all the older (1946 - 1950) samples, from which she reported the early parasitism of F. crotonensis, show the sole presence of rod-type diatom filaments. Illustrations, similar to rod-type and flared-type filaments of F. crotonensis were also presented by a number of authors in the early literature (see CANTER & JAWORSKI, 1982).

In the present study, however, F. crotonensis was represented only by rod-type filaments.

Parasite of F. crotonensis

Thallus, epibotic and monocentric. The sessile sporangium is spherical (Figs 39g & 40i-l) varying greatly in size, measuring

Fig.39. Fungal infection of Fragilaria crotonensis

a general appearance of fungal infection

b encysted zoospores

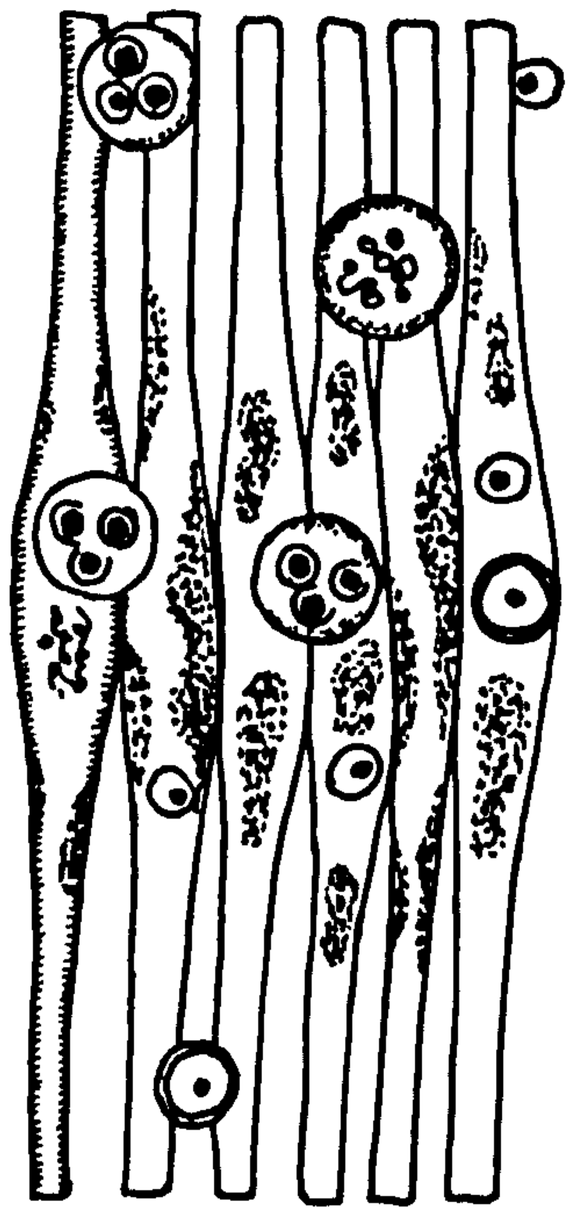
c developing sporangia

d - g mature sporangia

i empty sporangia

j resting spores

All drawings at X450



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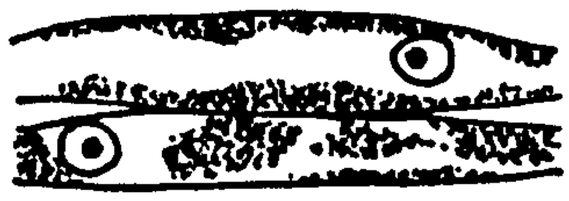

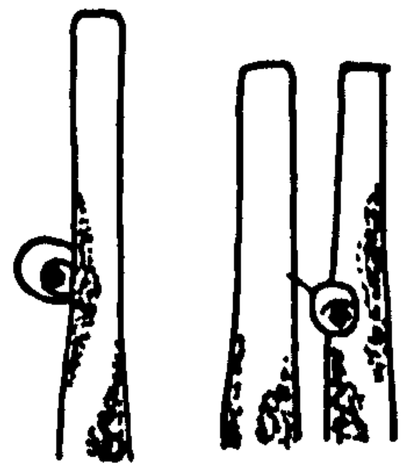

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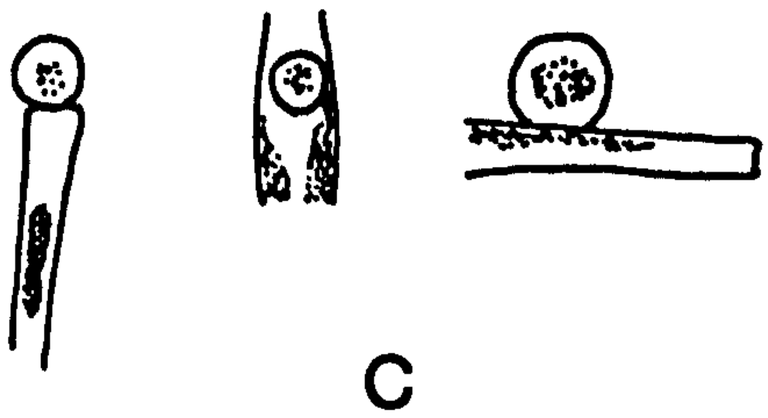

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d

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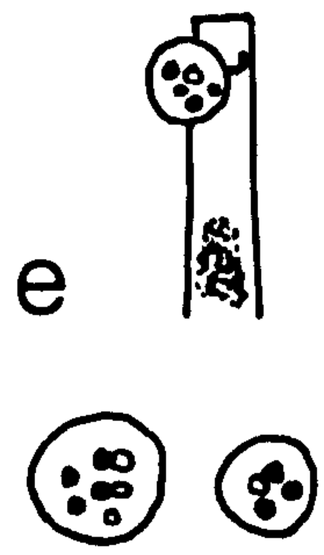

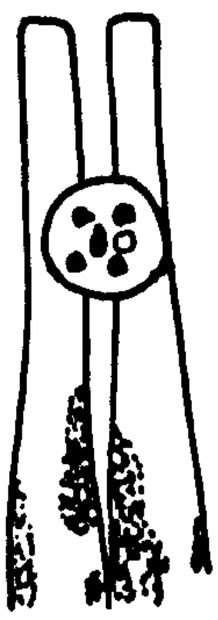

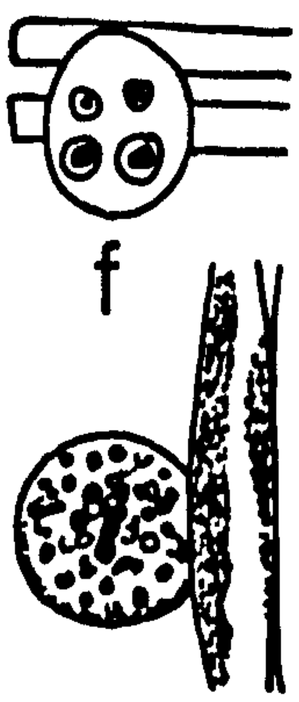

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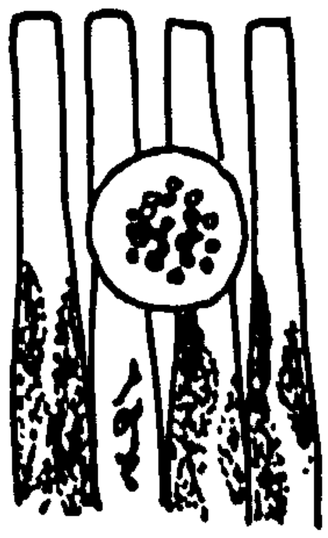
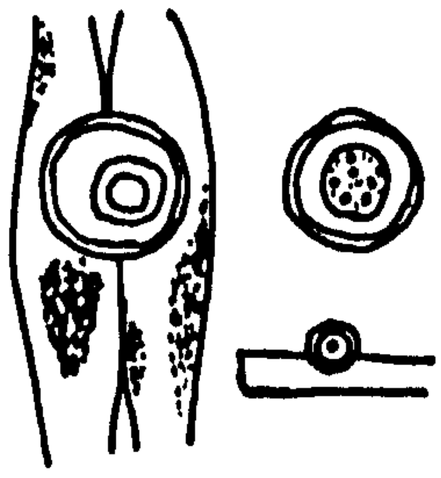


g

i



j

10µm

4 to 12.5µ in diam.,and occasionally somewhat oval in shape (Fig.39d,e). 9µ - 11µ long by 5µ - 6µ broad. Small sporangia produce three large oil globules (Fig. 39e) while the largest may contain as many as thirty small zoospores (Fig.39g,h). The intramatrical rhizoidal system was never observed. However, on a few occasions a short, thick unbranched single rhizoidal axis was observed (Fig.40f). Thus the sporangium of this fungus appeared to display more or less the same features as those of R. fragilariae and Z. affluens (parasite of A. formosa).

No data were obtained concerning dehiscence; this would have been a great help for identification. However empty sporangia (Fig. 29i) were rarely found on filaments which appeared to be rigid. The encysted zoospore is ovoid (Fig.29a,b) measuring 2 - 3µ long to 1.5 - 2.5µ broad. It contains a single oil globule and a nuclear cap is visible at some stages. Rarely a short germ-tube was also observed (Fig. 39b).

Resting spores were found during the most severe infection (September - October 1979). These were spherical, 4 - 9µ in dia., with a smooth wall. Internally it contains one or many small granules (Fig.39j). Resting spores appear morphologically to resemble those of Z. affluens and species 4. (see CANTER & LUND, 1953 p: 21-22). In addition the occurrence of the parasite of F. crotonensis sometimes coincided with the periods of parasitism of A. formosa by Z. affluens in this study, but at other times periods of infection on each diatom differed.

Further information was obtained on the fungal infection of F. crotonensis through a few occasional trips to Shearwater after the present investigation was terminated. Infection was recorded in November 1981 and details of the infection considered useful are therefore added to this section (Fig. 40). It is apparent that more developmental phases occurred during this infection. Germinating zoospores with or without a germ tube (Fig.40b-e)were quite abundant on filaments. Spherical, oblate and more or less oval sporangia, were all situated very close to the point of penetration into the diatom cell, (Fig.40g-l). Sporangia of R. fragilariae and species 3 can appear as more or less identical spheres when attached to Fragilaria filaments (CANTER & JAWORSKI, 1982). On a few occasions, sporangia with an externally placed thread (Fig.40f) were also observed. CANTER & JAWORSKI (1982) assigned this feature to species 3 as a distinguishing difference between R. fragilariae and species 3. More information was acquired on empty sporangia. Empty sporangia with a couple of openings (Fig.40p) characteristic of R. fragilariae (CANTER, 1950; CANTER & JAWORSKI, 1982) were found alongside the empty sporangia which appeared to be more or less similar to that of species 3. The latter sporangium varied in shape and often a few folds were found on the sporangial wall (Fig.40r). In addition, an operculum, almost triangular in shape (Fig.40r) was rarely found near the empty sporangia. This is also one of the features of species 3 (CANTER & JAWORSKI, 1982).

An endobiotic rhizoidal system was observed occasionally which consists of a conspicuous unbranched thread (Fig.40e). In this respect the fungus once more appeared to be similar to species 3. Lateral branches were not observed.

Fig.40 Light micrographs of fungal infection of Fragilaria crotonensis.

a - e germinating zoospores

d - h immature sporangia

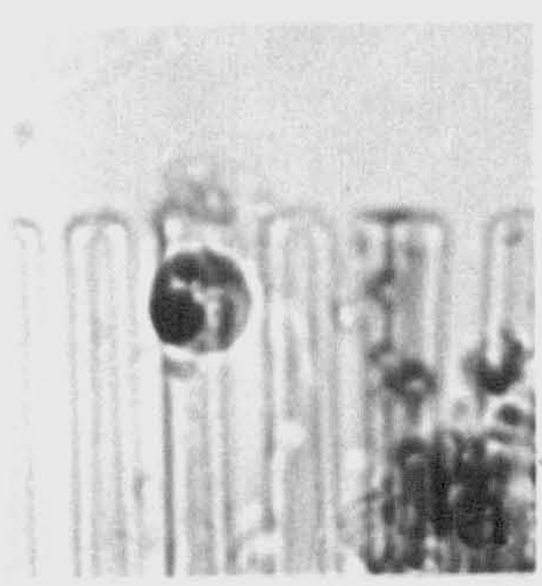
i - l mature sporangia

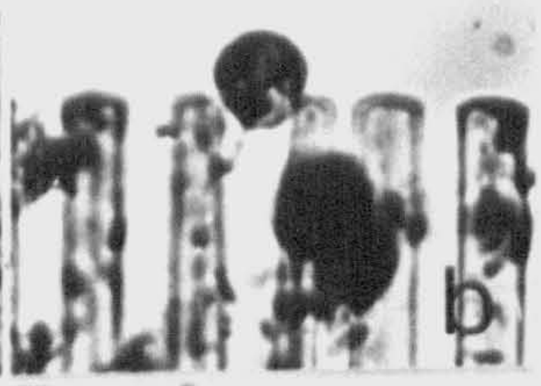
m - r empty sporangia

(→) indicates the lid (n), two openings (p) and triangular operculum (r) of empty sporangium

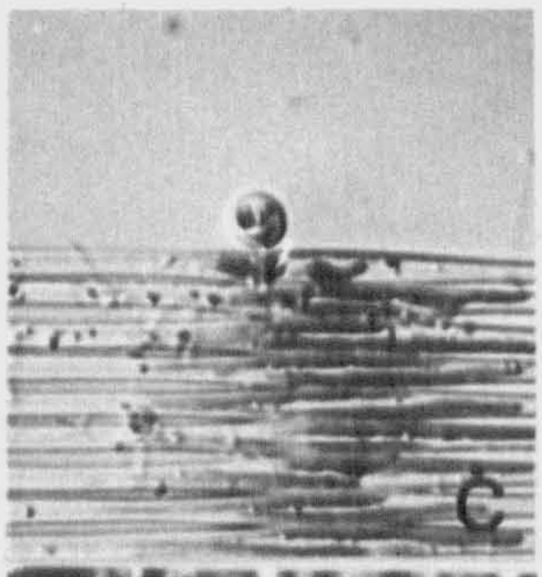
c, i, k, l, g at X450

The remainder at X860





b



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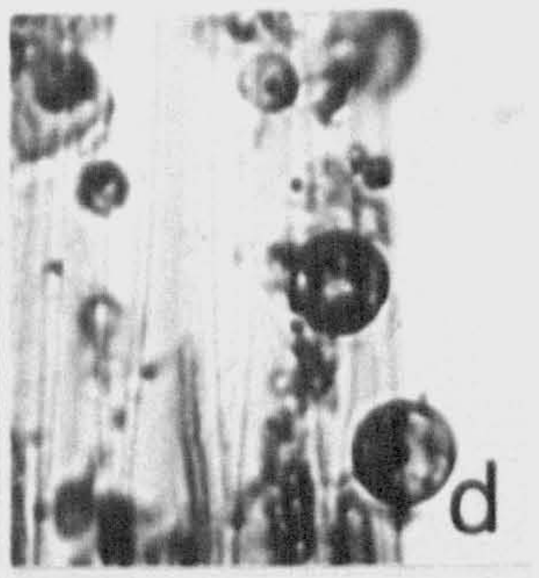

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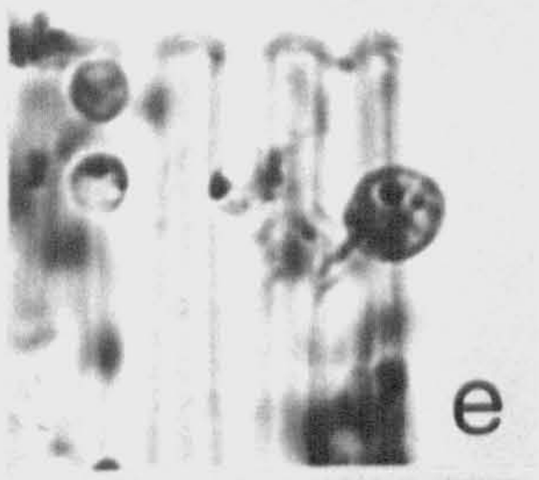

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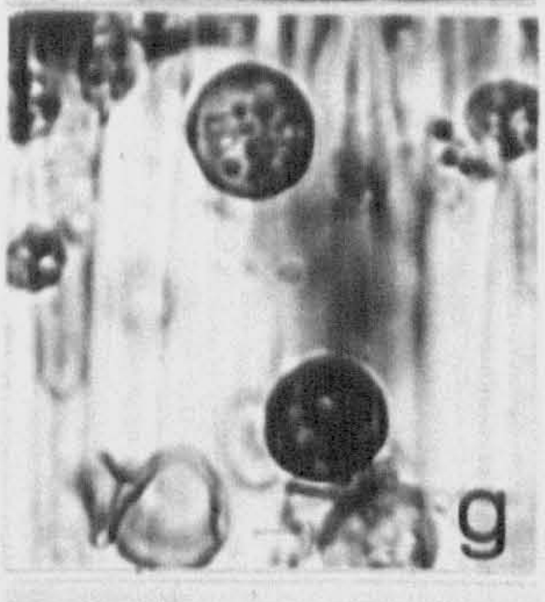

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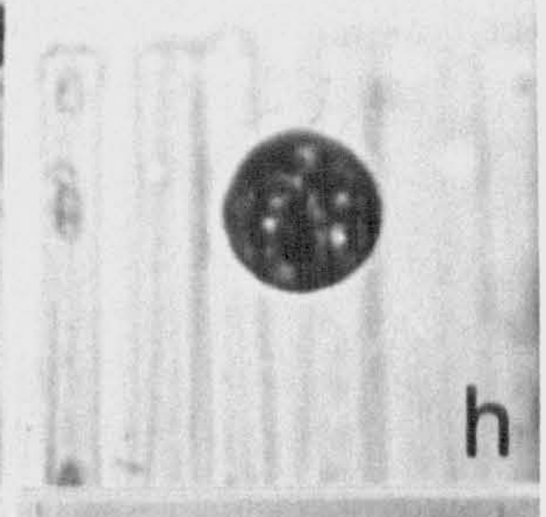

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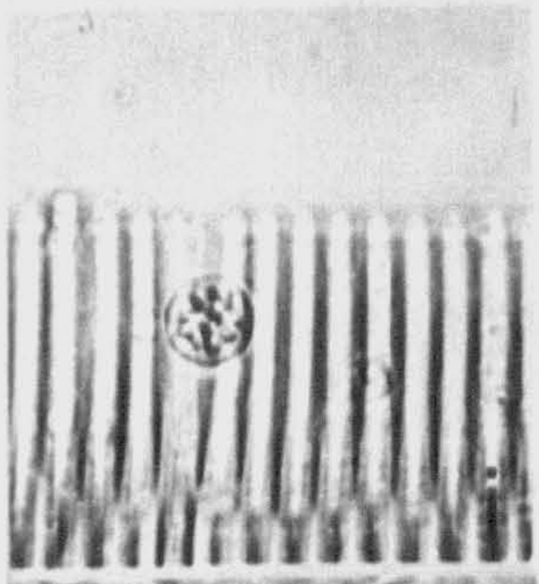
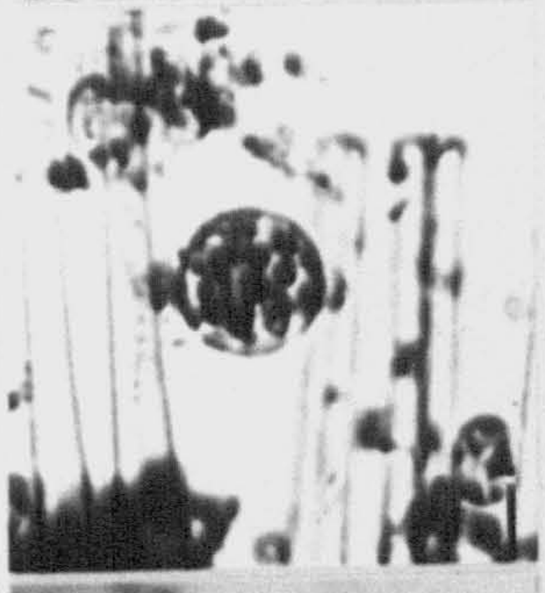
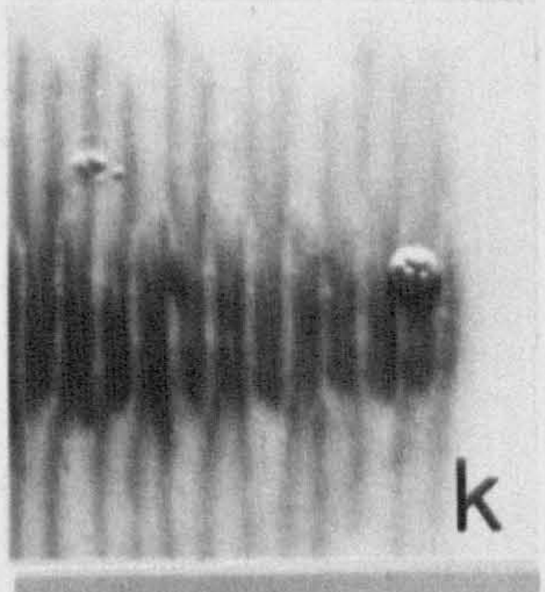
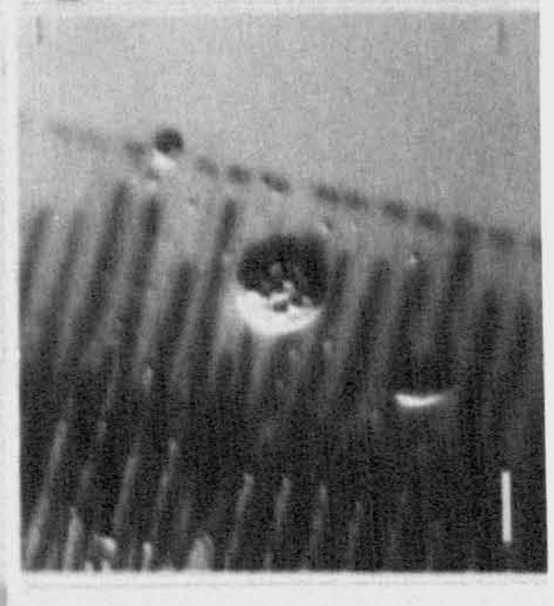


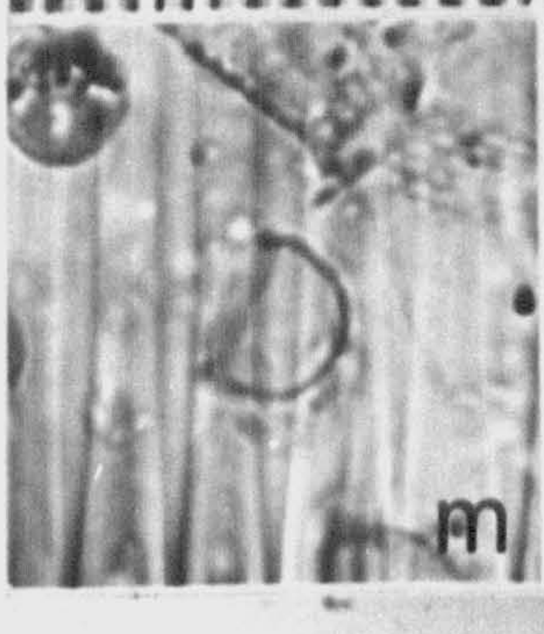




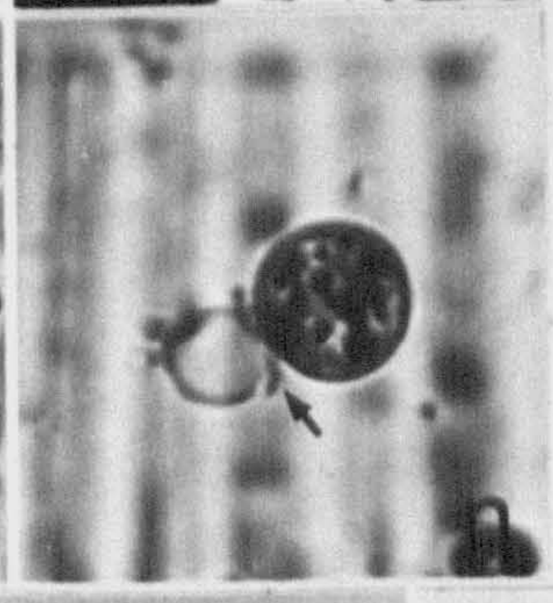
k



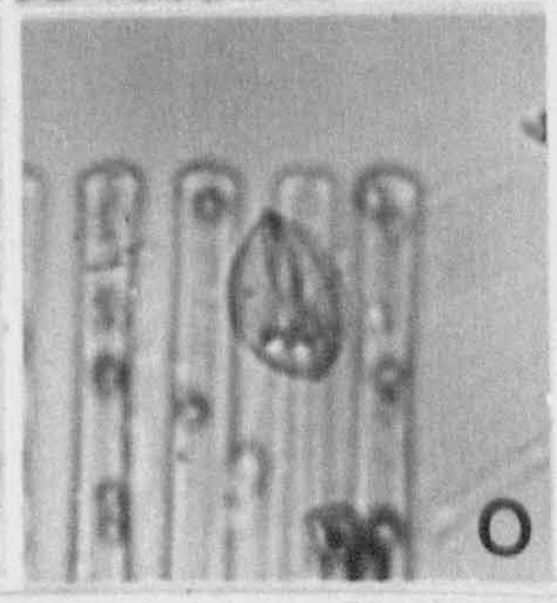
l



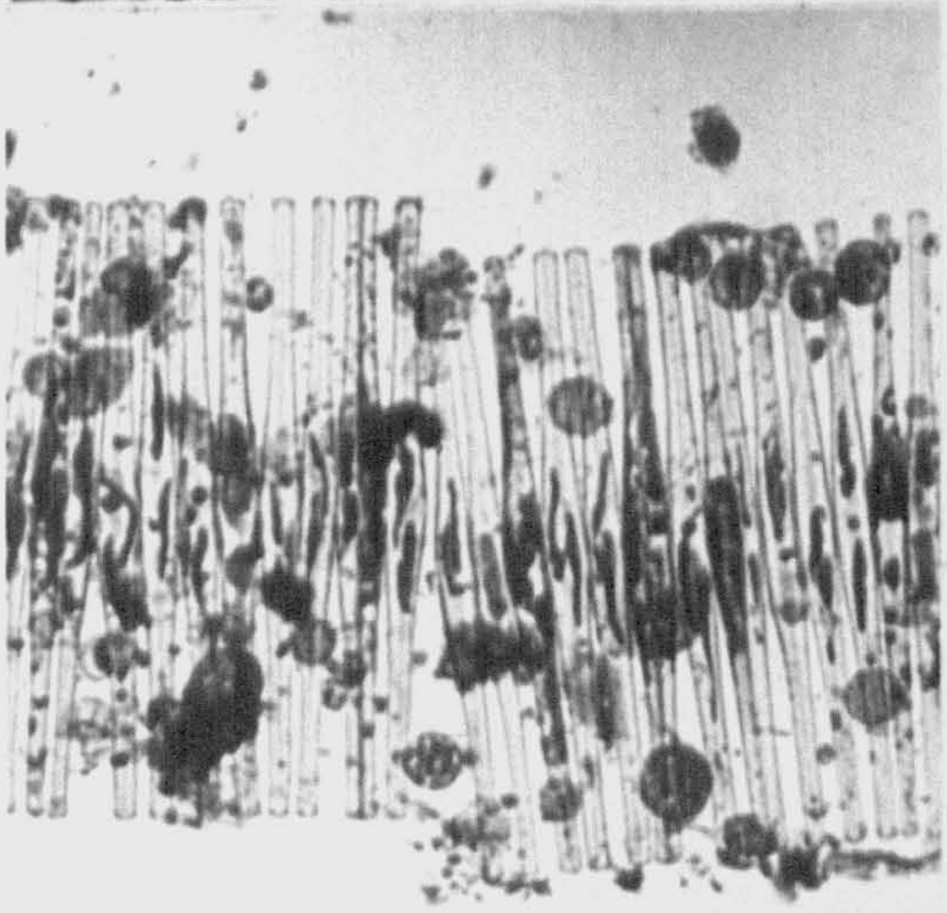
m



n



o



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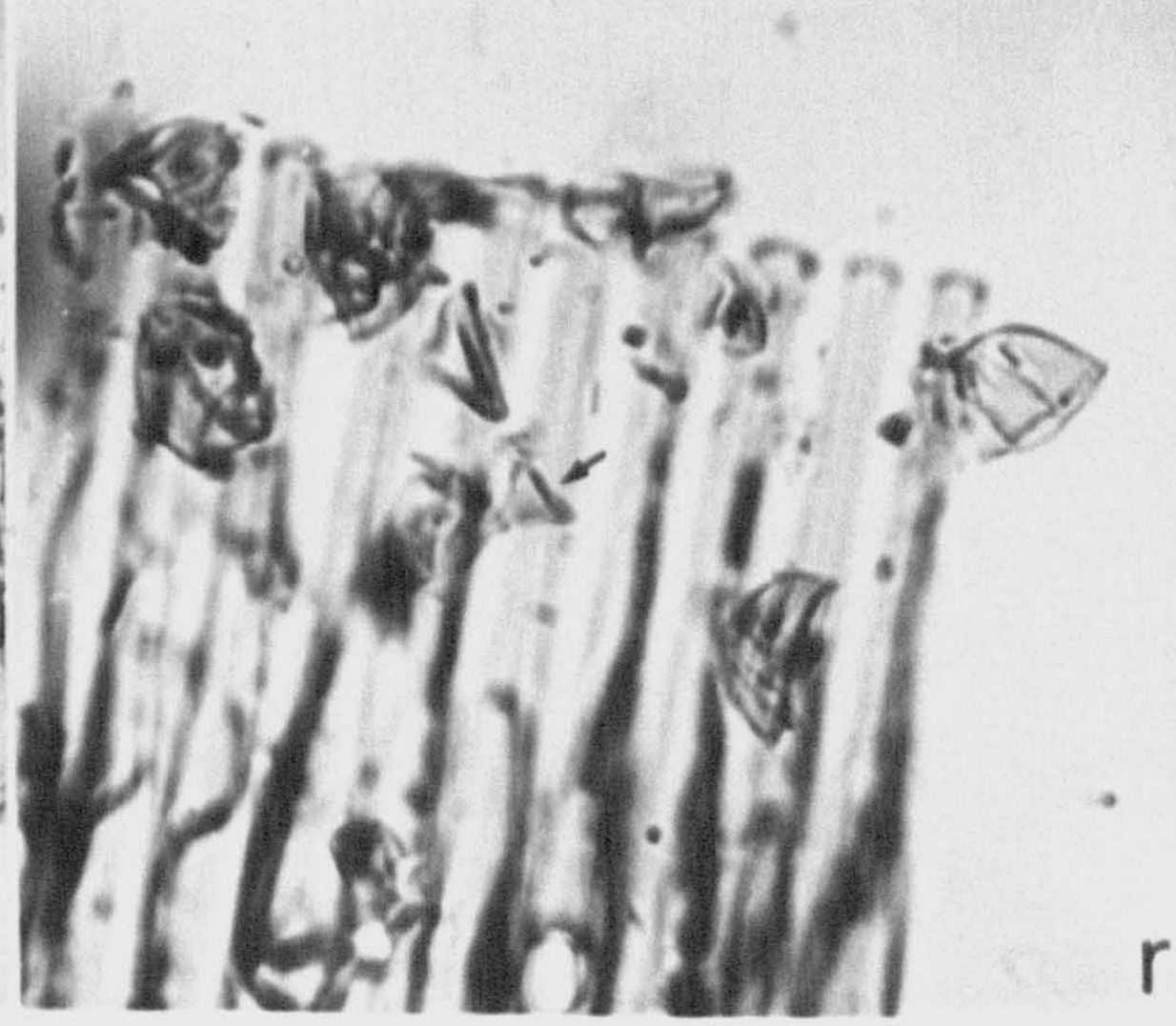


r

This additional information gave a chance to reach some conclusions on the fungal infection of F. crotonensis. Concerning all the features of the fungus during the present investigation, the fungus overall displays the characteristic features of both R. fragilariae and species 3 but not of Z. affluens. This might suggest that F. crotonensis may be parasitized by these two fungi in the present study. However it must be remembered that fungal epidemics on Fragilaria consisted mainly of encysted zoospores and mature sporangia during this investigation, thus which fungus caused the epidemics must remain in doubt.

Parasitism

The fungus tends to be a parasite since its development started on healthy and growing population of F. crotonensis.

Encysted zoospores were most frequently found on healthy cells. In healthy cells, chromotophores occupy a large proportion of the cell body (see healthy cells). A few cells, infected by encysted zoospores nevertheless appeared to be unhealthy, probably due to additional bacterial infection also (Fig.40d). The cells, bearing sporangia and resting spores were mostly dead although some still appeared to be healthy (Fig.39g,j). Chromotophores were very much reduced in dead cells and in some cases granular remains were characteristic. It is noteworthy that there was also bacteria infection on the cells in the periods of fungal epidemics, thus maybe masking the effect of fungal parasitism on the cells. Because of this

the increase in the number of dead cells which was recorded during fungal epidemics could not be shown quantitatively. However, CANTER & LUND (1953) reported sharp declines in the average number of live cells per filament (e.g. from 69 to 3.2) during fungal parasitism of F. crotonensis.

Nevertheless, developmental phases of the fungus during epidemics is shown in Fig. 41. Epidemics of this fungus, in this respect, displayed a few features not shown by other infections of phytoplankton in Shearwater.

Fig. 41 clearly shows that fungal infection of Fragilaria consisted dominantly of mature sporangia. However short-term epidemics with low infection rate (2nd April 1979; 20th May, 8th December 1980) were characterised by encysted zoospores. Due to an unknown factor encysted zoospores did not develop into sporangia, on the contrary, they just disappeared after a fortnight. Summer epidemics (June 1979; June-July 1980) started in an unusual way with dominant numbers of mature sporangia, and the first epidemic ended in the same way. During the last stage of the second summer epidemic, sporangia were still dominant whilst few encysted zoospores or developing sporangia were found. The onset of the longest and most severe epidemic (September 1979 - February 1980) coincided with high numbers of resting spores and low numbers of encysted zoospore and mature sporangia. This was quite in contrast to the onset of other fungal epidemics recorded in this study. At the stage of maximum infection, the numbers of encysted zoospores and mature sporangia were increasing while the numbers of resting spores were decreasing. At the next stage, mature sporangia became dominant in numbers and their dominance continued for

Fig.41. Developmental phases of the fungus on F. crotonensis during epidemics

[X] encysted zoospores

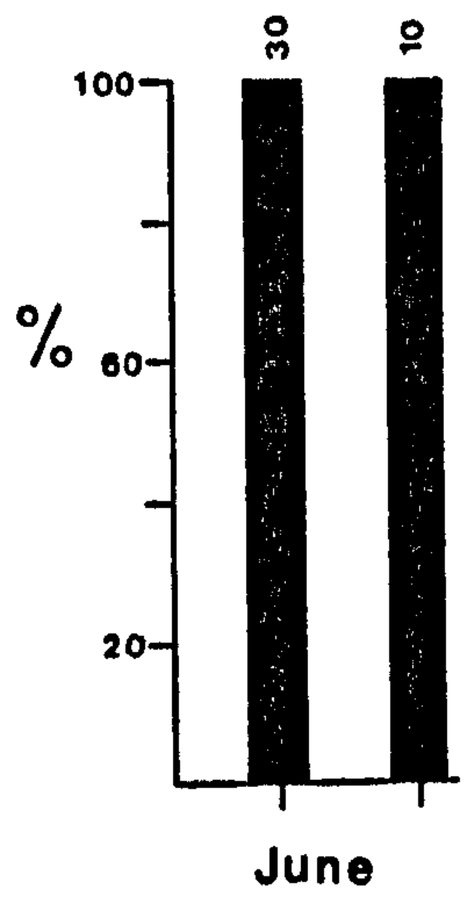
[II] developing sporangium

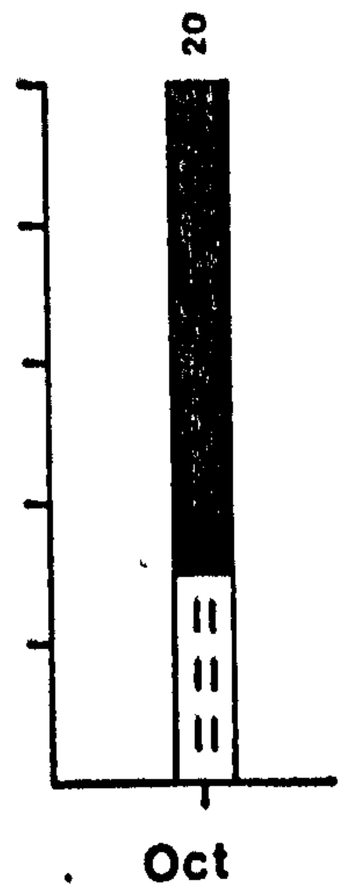
[■] mature sporangium

[O] empty sporangium

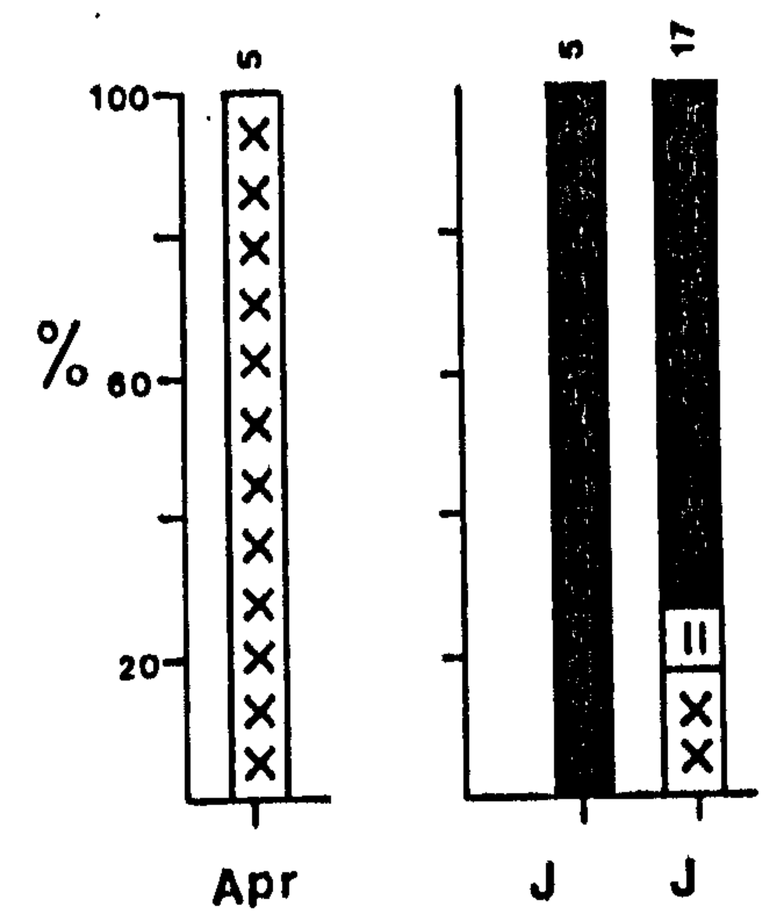
[ ] resting spore

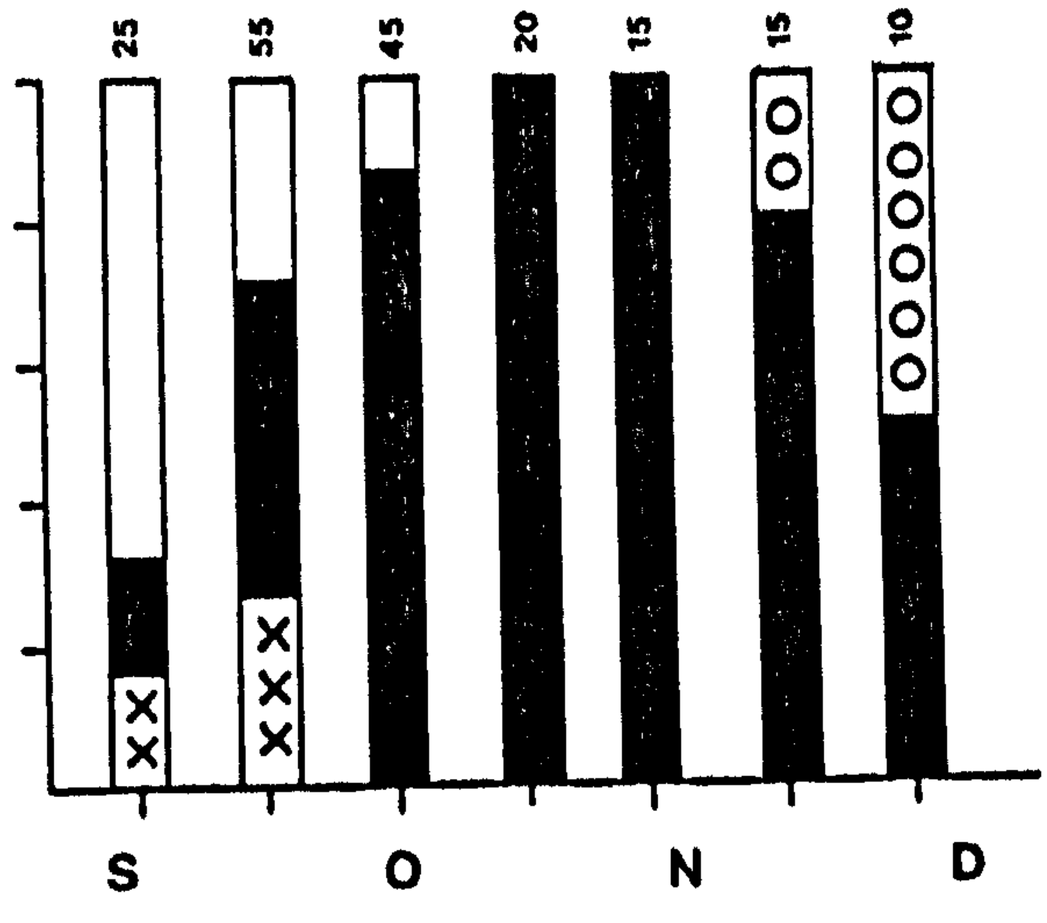
Note: Numbers indicate the % fungal infection



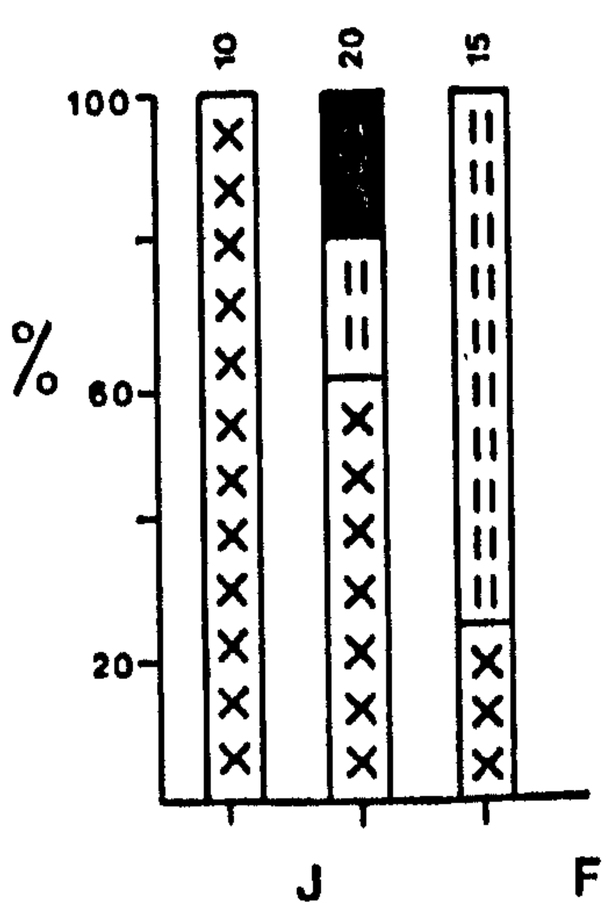


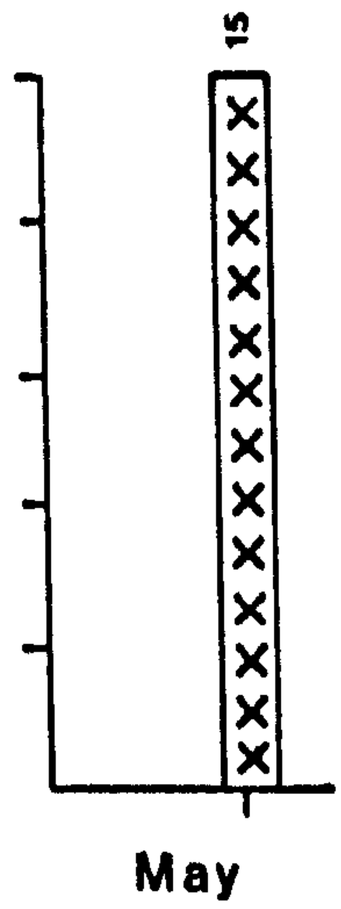
1978

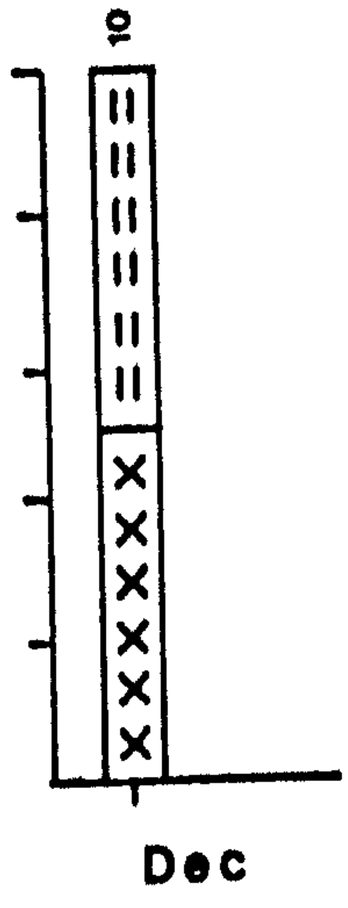




1979







1980

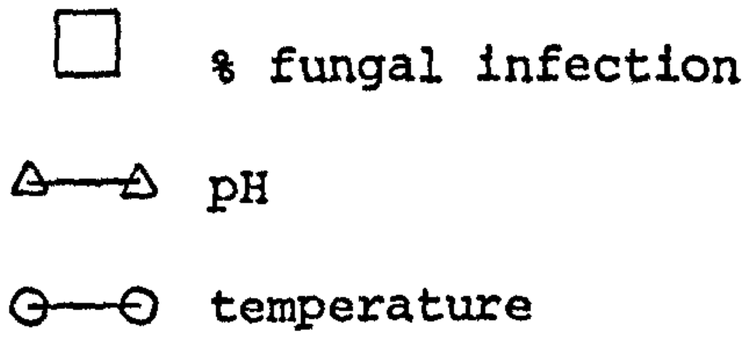
almost three months. Empty sporangia were found only during this severe epidemic long after the epidemic started. The next stage of the epidemic was quite interesting in that encysted zoospores became dominant on the cells. This was probably connected with the presence of empty sporangia at the previous stage, indicating the release of new zoospores. Next, the numbers of developing and mature sporangia increased while encysted zoospores decreased in number. It is noteworthy that there was an increase in the degree of infection as well. The epidemic finally ended with increased number of developing sporangia. This might suggest that a second development of the fungus started within this long epidemic and also suggests that development of the fungus possibly could continue stage after stage if the conditions were favourable.

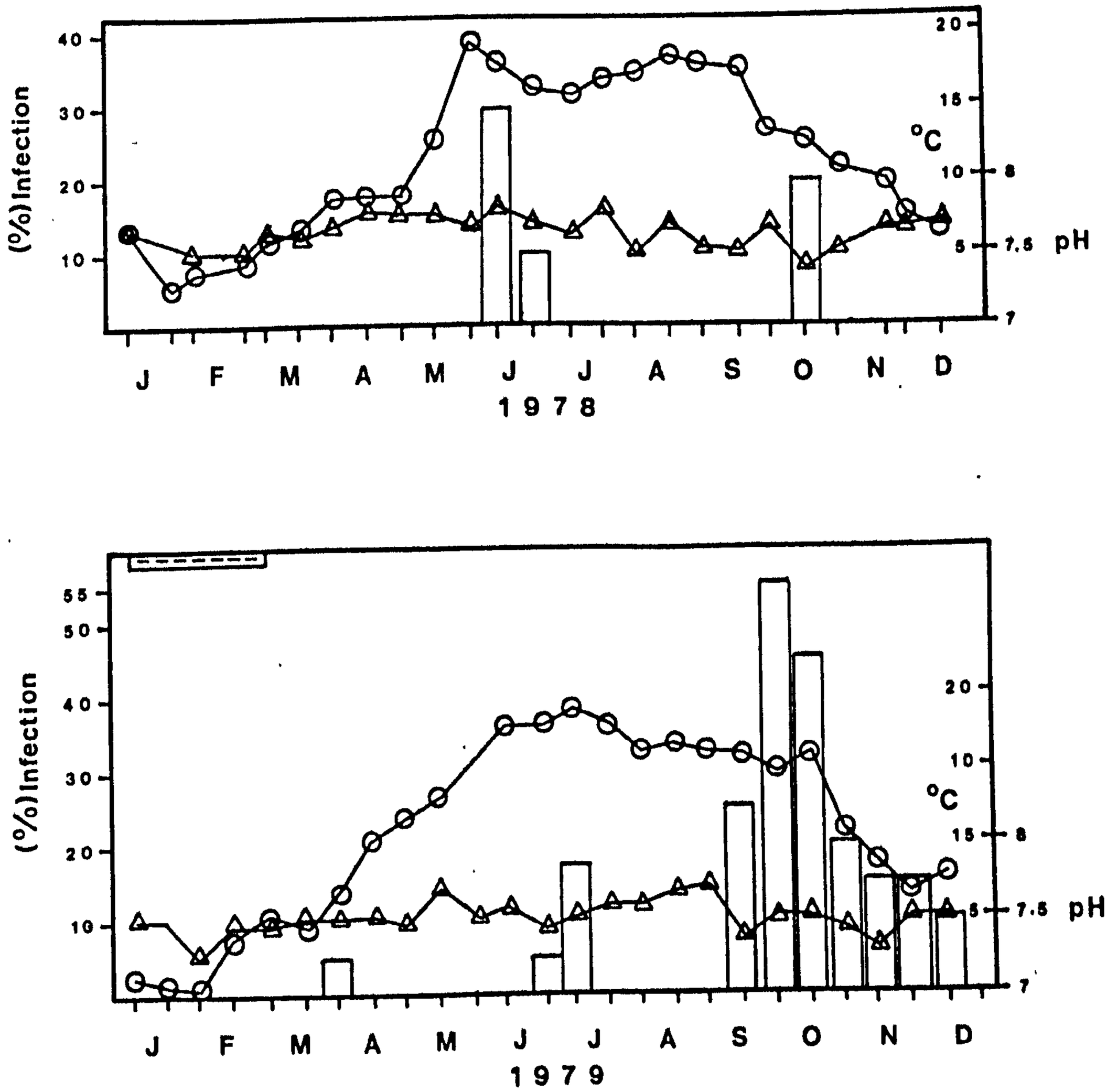
Consequently, epidemics of this fungus, in this respect, had different features. Dominant numbers of mature sporangia and resting spores at the onset of the epidemic was quite in contrast to epidemics of other chytrids, which usually started with high numbers of encysted zoospore and developing sporangia and ended with sporangia being dominant.

Epidemics

Epidemics of the fungus are shown in Fig. 42 in relation to physical factors which can be compared with the fluctuations in the number of Fragilaria (Fig. 7) and chemical data (Fig. 3). It is apparent from the Fig. 42 that the occurrence of the fungus was irregular since it occurred in different years at

Fig.42. Fungal infection of F. crotonensis in relation to physical factors.




(%) Infection
40
30
20
10
°C
20
15
10
8
7.5
7
pH
J F M A M J J A S O N D
1978
55
50
40
30
20
10
1979
NO DATA
1980

different seasons.

In 1978, the first epidemic was recorded on 11th June when 30% of the cells were infected. The growth of the host population before the epidemic reached 504 fils/ml. After a fortnight infection declined to 10% and Fragilaria reached a maximum of 2149 fils/ml. After this epidemic, the diatom decreased sharply apparently because of exhausted silica supply. In the same year another epidemic occurred around the 16th October with a slightly lesser degree of infection (20%). Fragilaria was increasing before the infection and a sharp declination in numbers occurred (from 396 to 88 fils/ml.) with the onset of the epidemic. This seemed also to be connected with the reduction of silica concentration to 0.1 mg/l (Fig. 3). The fungus was absent a fortnight later.

In 1979, the first appearance of the fungus was recorded on 2nd April with very low infection level (5%) at a time when Fragilaria colonies were declining rapidly in the water column (down from 110 at the previous collection date to 17 cols/ml). However this decline may not be attributable to this low fungal infection but may be to decreasing silica concentration (Fig. 7). It is noteworthy that the diatom increased in numbers after this very short appearance of the fungus with the support of increasing silica. A more severe infection was recorded in summer (27th June - 9th July). The epidemic started with 5% infection and increased to 17% after a fortnight. Increasing host population before the onset of epidemic declined to 50 fils/ml. when epidemic started and this was followed by a slight increase in the numbers by the maximum infection. Silica level

was rising during this period but it was still at low concentration. The diatom population was decreasing after the epidemic although the silica level was rising sharply. A. formosa was in its period of active growth and this appeared to be connected with declination of Fragilaria population perhaps as the two diatoms competed for silica. The numbers of Fragilaria continued to decline under the pressure of active growth of Asterionella, until the onset of the longest and the most severe infection ever recorded on Fragilaria during this study. In fact, this period of epidemic was the longest of all the fungal epidemics recorded on the phytoplankters of Shearwater in the present study. It lasted almost five months.

This major epidemic started in September (17th) with an infection degree of 25% and reached a maximum infection of 55% after a fortnight. After this stage,infection level started decreasing and continued to decline gradually until 7th January 1980 (down to 10%). By 21st January 1980 the infection had suddenly increased to 20% and a slightly lesser infection (15%) was recorded subsequently on 4th February. After a fortnight the fungal infection disappeared from Fragilaria although high numbers of filaments were still available in Shearwater.

In spite of this lengthy infection, fluctuations in the numbers of Fragilaria showed only a slight correlation with parasitism. There was a slight increase in host population before epidemic started. The maximum stage of the infection (September - October 1979) coincided with a slight declination of host population. However, the decrease (from 52 to 14 fils./ml.) was not as consideable as might have been expected during such a severe infection. The effect of parasitism on the

decrease of the host population was also masked by the effect of decreasing level of silica. Asterionella was also decreasing in numbers in the same period, probably due to decreasing silica level, although there was no fungal infection on the cells. After the maximum of fungal infection until the end of the epidemic (November 1979 - February 1980), the host population fluctuated sharply although the infection level was more or less stable (varying between 10% - 20%). Concentration of silica was generally increasing during this period thus favouring diatom growth. This may suggest that some other factors alongside with parasitism and silica might also be in charge of governing the seasonal periodicity of F. crotonensis. A slight decrease in the number of Fragilaria was recorded after the epidemic ceased.

In 1980, two more short appearances of the fungus were also recorded, on 20th May and 8th December respectively. In both cases, the fungus had disappeared by the next sampling date. The numbers of Fragilaria were increasing during the first infection while a slight decrease was recorded on 8th December, although the former infection (20%) was more severe than the latter (10%).

It became quite clear that most fungal epidemics occurred in the periods of active growth of Fragilaria. However at one stage occurrence of an epidemic (June 1979) coincided with the period when the host population was about to decline from other causes thus supporting the view of CANTER & LUND (1951). No renewed increase in numbers of Fragilaria occurred after thiis epidemic.

Effects of parasitism on the numbers of Fragilaria appeared to be obscure. Sharp declines in the cell number of the host population did not really occur unlike the case of parasitism of Asterionella by Z. affluens in this study. But low silica values were always masking the effect of parasitism of Fragilaria. In addition, even during the most severe and longest epidemic, the decrease of the host population was very small and numbers seemed to increase or decrease regardless of the parasitism. However CANTER & LUND (1953) found that parasitism increased the rate of decrease in the number of F. crotonensis in the English Lake District.

In several cases, however, parasitism of F. crotonensis occurred at a time when particular chemical conditions were unfavourable for the growth of the diatom and its growth might have been stressed by these factors. Nevertheless, the fungus did not seem to enhance the decline of the diatom.

One of the epidemics had a feature which agrees with the view of CANTER & LUND (1951) that parasitism may delay the time of the algal maximum; it may also decrease its size. In the present study, Fragilaria reached its autumn maximum either in October or November in three years (1977, 1978, 1980); during these periods slight fungal infections or none were recorded. The sizes of autumn maxima varied between 300 to 600 fils./ml. in these years. However,during autumn in 1979, the longest and the most severe epidemic occurred and the autumn maximum of the diatom was late occurring (in December). In addition, its size was smaller (239 fils./ml.) than those of other years.

It is noteworthy that summer maxima of Fragilaria always occurred in June over the period studied whether there was an infection or not on the diatom. In addition the summer maximum in 1978 was the largest (2148 fils./ml) although 30% of infection was recorded on Fragilaria.

In conclusion, it became clear that seasonal periodicity of F. crotonensis cannot be interpreted without reference to parasitism although its effect is not as extreme as that of the parasitism of other planktonic algae.

Fungal infection and frustule lengths of the cells of F. crotonensis.

Differing frustule lengths of Fragilaria also appeared to be of some interest in the case of fungal parasitism of this diatom as was also recorded for A. formosa in this study.

The temporal variation in frustule length of F. crotonensis is well known from the beginning of this century. SCHRÖTER & VOGLER (1901) recognised four discontinuous size classes of the diatom in the lake of Zurich. They designated curta of modal length 57 to 60µ, media of modal length about 78µ, subprolongata of modal length about 104µ, and prolongata of modal length 126 to 129µ. In the study of WESENBERG-LUND (1904), two genuinely distinct populations of Fragilaria were present. One was undergoing reduction in size from 140 to 133µ over a period of about a year while the mode of the other was quite stationary over the same period at 112µ.

Frustule lengths of Fragilaria were shorter in this study compared with the ones recorded by the above authors.

Measured frustule lengths of Fragilaria varied between 65µ - 84µ during this study. Table 7 shows the combination of frustule lengths occurring in Shearwater and the comparison of fungal infection of these frustules.

Frustule length	65µ	68µ	71µ	73µ	76µ	78µ	81µ	84µ
Total Cells Counted	35	140	600	1475	1705	1100	190	45
% P. infection	-	5%	7%	29%	40%	12%	6%	1%

Table 7. Comparison of parasitism of frustule lengths of F. crotonensis.

As is seen from the above table frustules shorter than 71µ and longer than 78µ were relatively scarce. The frustules of 73µ and 78µ length were dominant in the population of Fragilaria throughout this investigation. This result would suggest the occurrence of only one Fragilaria population in this study.

Degree of fungal infection paralleled the numbers of frustule lengths in the present study.

It is apparent from Table that the frustules 73µ and 76µ in length succumbed to fungal attacks far more frequently than other frustule lengths, probably due to their dominance in the Fragilaria population. It might be considered obvious that whichever frustule length was most frequent, the greater the infection might be on these cells, considering that this fungus grows only on Fragilaria. Searching throughout the literature, no references were encountered concerning the distinct frustule lengths of Fragilaria in relation to parasitism.

The onset of epidemics in relation to physical-chemical factors

Of all the physical-chemical factors studied, no single nor group of factors could be demonstrated to determine the occurrence of an epidemic. The following factors have been considered with inconclusive results.

Temperature: occurrence of the fungus coincided with high temperatures as well as low temperatures in the present study (Fig. 42). Concerning the epidemics with high degree of infection, high temperatures appeared to favour the growth of the fungus. Four epidemics occurred within the temperature range 15°C - 20°C. In addition, the most severe stage of the longest epidemic was synchronous with a temperature of around 15°C. However, an increase in the degree of infection was also recorded under the ice when the temperature was only 3°C although low fungal infections were generally recorded at low temperatures (5 - 8°C).

pH: variations in pH level were generally slight in Shearwater, thus it might be considered to have less effect on the growth of fungus than other factors. Apart from the epidemic recorded June 1978 (pH 7.7 - 7.8), the rest occurred within the pH level of 7.4 - 7.5 (Fig. 42), therefore one might assume that pH 7.4 - 7.5 might be most suitable for this fungus.

Water level: the onset of epidemics often synchronized with rising water level (Fig.2). However the percentage infection during epidemics did not show a striking correlation with the rise and fall of water level.

Nitrate: concentrations of nitrate were usually high or rising when epidemics started (Fig. 3). The lowest concentration was 0.26 mg/L. Thus this might suggest that concentrations below 0.26 mg./L were not favourable for the growth of the fungus.

Phosphate: high or low concentrations of phosphate coincided with the onsets of epidemics (Fig. 3). The changes in phosphate level were not paralleled by those in degree of infection.

Silicate: all but one epidemic occurred when silica was already at high levels or rising (Fig.3). However the onset of an epidemic (16th October 1978) with an infection degree of 20% was synchronous with the lowest concentration of silica (0.1 mg./L) recorded during this study.

Abundance of F. crotonensis: abundance of the diatom did not correlate with the degree of infection in this study. Epidemics occurred in the periods of either low or high numbers of Fragilaria. For example, the epidemic (30% infection) in July 1978 occurred when Fragilaria was very close to its maximum (504 fils./ml.) while the longest and the most severe epidemic (55% infection) started when the diatom was lower in numbers (52 fils./ml.). However all epidemics occurred when Fragilaria was present over 50 fils,/ml., exclusive of a low infection on 2nd April 1979 (5% infection) when the number of Fragilaria was only 17 fils./ml. This might suggest that the lower limit of Fragilaria may be 50 fils./ml. for the fungus to reach the epidemic proportions (over 20% infection).

Summary of Conclusions

Fragilaria crotonensis was represented only by rod-type filaments which may possibly be infected by two morphologically similar chytrids: Rhizophydium fragilariae Canter and species 3.

The occurrence of epidemics on Fragilaria was irregular and usually coincided with the periods of active growth of the diatom. However at one stage an epidemic was synchronous with declining host population from other causes.

During the longest and the most severe epidemic development of a second attack may indicate that the development of chytrids can continue stage after stage for a long time if the conditions are favourable.

No single or groups of factors could be demonstrated for the occurrence of the epidemics.

At least 50 fils./ml. of Fragilaria appears to be a limit for the fungus to reach epidemic proportions.

Distinct frustule lengths of Fragilaria were present and some were infected more than others.

Fungal infections consisted mainly of encysted zoospores and mature sporangia.

Epidemics usually commenced with high numbers of mature sporangia and the most severe epidemic with dominant numbers of resting spores. This feature contrasts with other fungal epidemics, recorded on other algae in this study which start with high numbers of encysted zoospores and terminate with dominant numbers of sporangia.

Empty sporangia were found during only the most severe and the longest epidemic.

Highest infection rate was 55%.

The effects of parasitism on the numbers of Fragilaria remained obscure since its effect is not as extreme as that of parasitism of Asterionella.

Ratio of dead cells to live cells increased during all the epidemics.

The autumn maxima of Fragilaria was later and smaller in size during an epidemic than at other times when infections were not present.

Chytrid infection of other diatoms

Apart from the severe parasitism of Asterionella formosa and Fragilaria crotonensis by chytrids, the fungal infection on other diatoms remained relatively unimportant in Shearwater.

Centric diatoms and fungal infection

Centric diatoms were common members of the phytoplankton of Shearwater, particularly Cyclotella spp. and Stephanodiscus hantzchii; they occurred in high numbers in autumn and spring. Fungal infection on centric diatoms was, however, relatively low despite their striking numbers.

Fungal parasite of Cyclotella spp.

The identification of infected Cyclotella spp. was quite difficult since the observations were carried out on live materials. However a spherical epibiotic chytrid was found on

one or more of the following: Cyclotella meneghiniana, C. kutzingiana in Shearwater.

Spherical or ovate encysted zoospores, 1µ to 3µ in dia., or height were observed to be attached to the cells (Fig.43a-c). They contain a single oil globule and a nuclear cap was visible on most occasions (Fig. 43a). No germ tube was seen. Young sporangia (Fig. 43f) were more or less spherical and internally contained numerous granules. Mature sporangia vary in shape from more or less spherical (Fig. 43i), 10 - 11µ in dia., to egg-shape (Fig. 43g), 7 - 8µ long by 5 - 6µ broad. Sporangia may contain 3 to 30 oil globules according to size. The sporangia appeared to be sessile. Neither a rhizoidal system or a stalk were observed. In addition no data were obtained on dehiscence and resting spore formation. Empty sporangia were scarcely found (Fig. 43j) on the cells,giving no evidence whether it is an operculate or inoperculate.

CANTER & LUND (1953) gave brief information on fungal parasitism of several Cyclotella spp. in samples taken from England and part of Europe. They also found a spherical, epibiotic chytrid, growing on several Cyclotella spp. It might be possible that the chytrid on Cyclotella spp. in this study might be the same chytrid, recorded by CANTER & LUND since they are both spherical. The same authors (1953) also observed a polyphagoid chytrid, internal fungi and a fungus resembling Lagenidium cyclotellae Scherffel (biflagellate fungus) on and in Cyclotella spp. L. cyclotellae appears to be the only fully described fungus on Cyclotella (originally described on C. kutzingiana). However the fungus in this study showed no resemblence to L. cyclotellae. Identification of the fungal

Fig.43. Fungal infection of Cyclotella

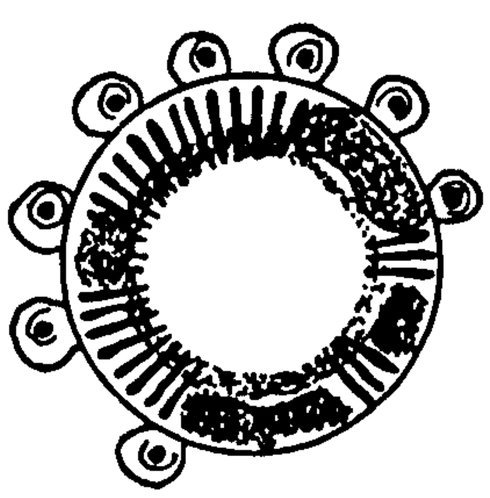
a - c encysted zoospores

d - f developing sporangia

g - h mature sporangia

j empty sporangia

all pictures at X450



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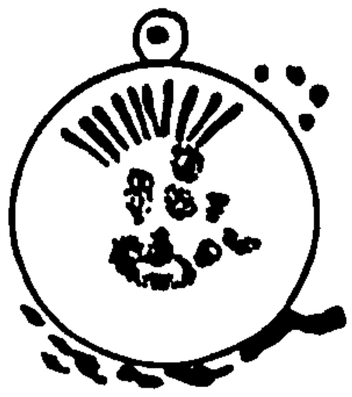

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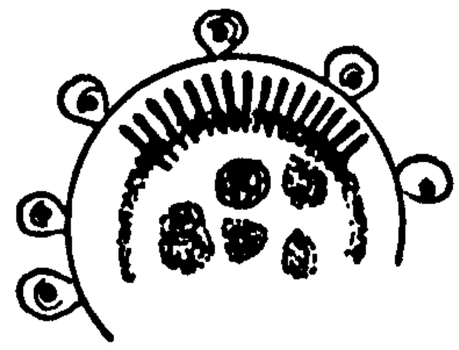

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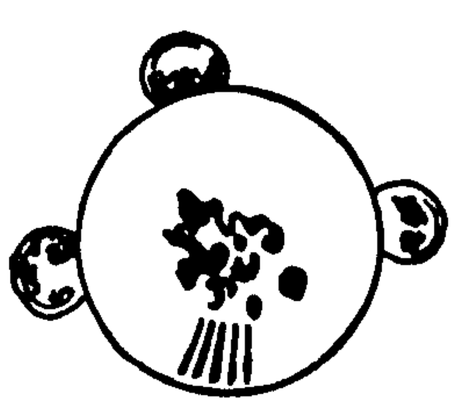

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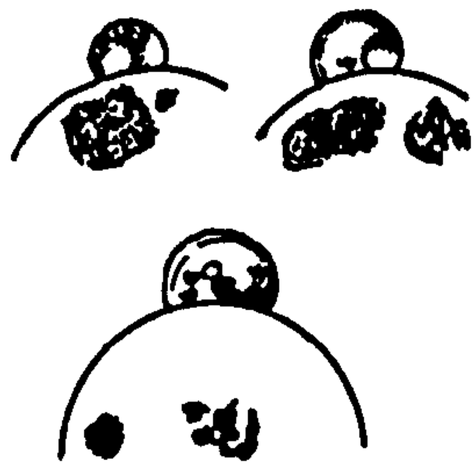

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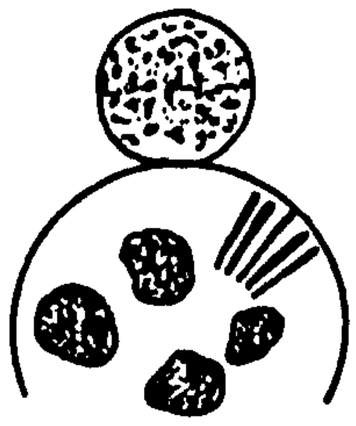

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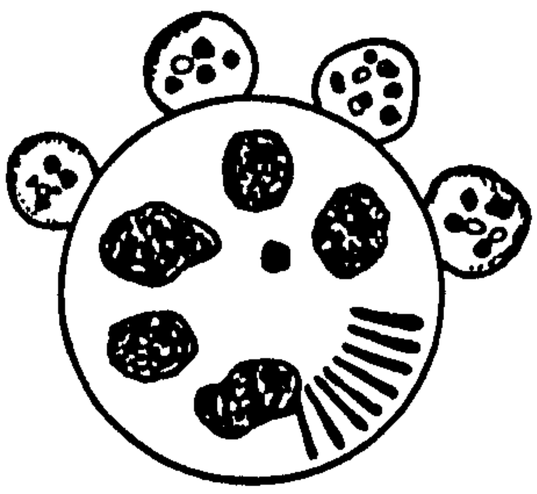

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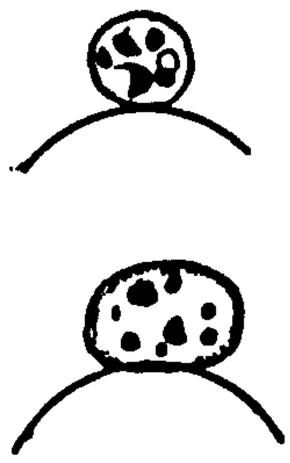

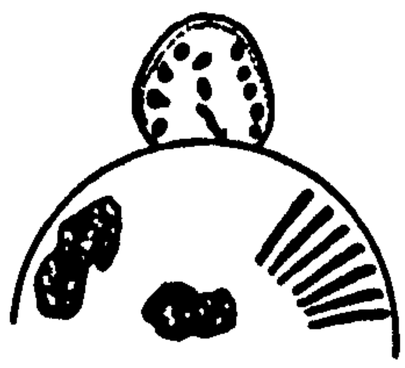

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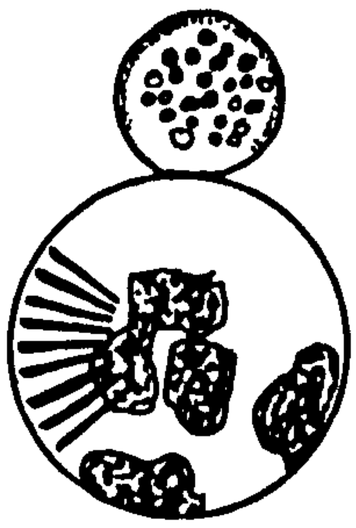

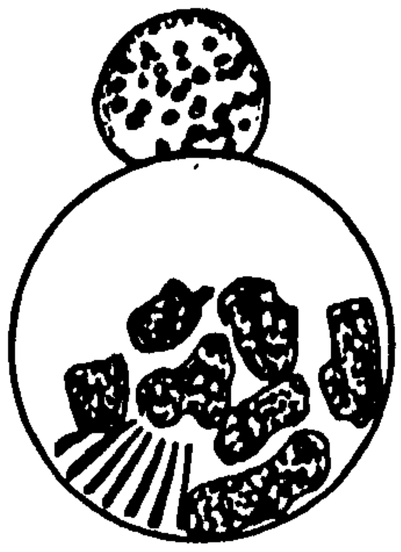

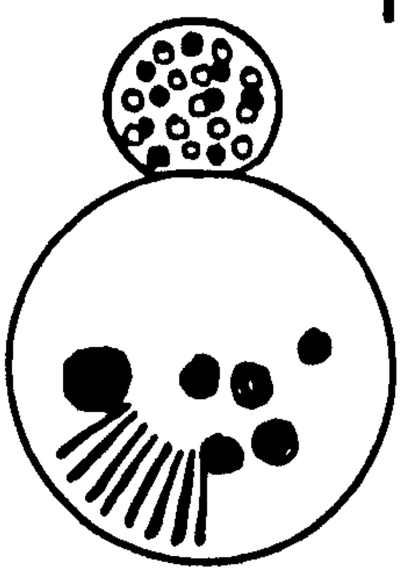

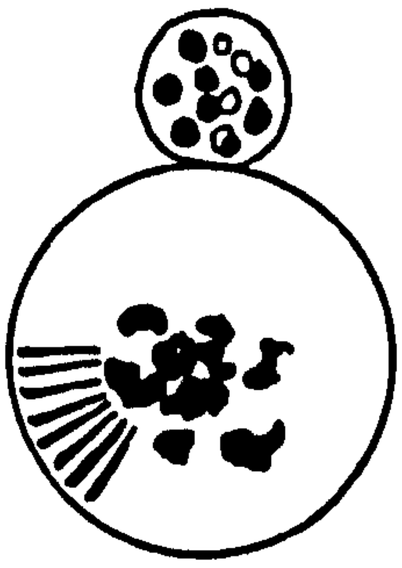

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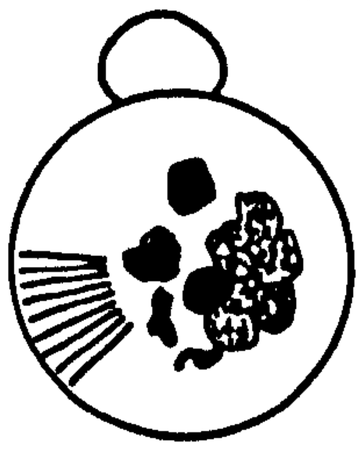


j

10µm

parasites of Cyclotella in this study and in other lakes certainly requires more information especially on full life cycles.

Fungus on Stephanodiscus hantzchii Grun.

S. hantzchii Grun. was the only member of the genus occurring in Shearwater. The cells were found to be rarely infected by an epibiotic fungus during this investigation.

Very little data was obtained on the fungal infection of S. hantzchii since the infection was very slight. Large encysted zoospores were oval in shape (Fig.44a-b),3 to 4, 5μ high by 1.5 - 3μ broad, may be seen attached to the cells. They contain a single oil globule and no germ tube was found. Encysted zoospores were always accompanied by sporangia on the cells. Sessile mature sporangia appeared to be more or less spherical (Fig. 44c,d)8μ in dia., containing approximately 15 - 20 oil globules. Two types of empty sporangia were observed. One had a spherical base and tapered distal end (Fig.44e,fmeasuring 15 - 16μ high to 10 - 11μ broad . It had a thick segment near the apex. The second type was bowl-shape (Fig.44h,j) 10 - 11μ broad. This type also had that characteristic thick segment near the upper portion. Thus it might be possible that these two empty sporangia are the same species but at different stages of growth. A thick dot-like process was also observed on the bases of both empty sporangia. In addition, at one stage, more or less a spherical empty sporangium with disconnected upper portion (Fig.44g) was also seen. Disconnected

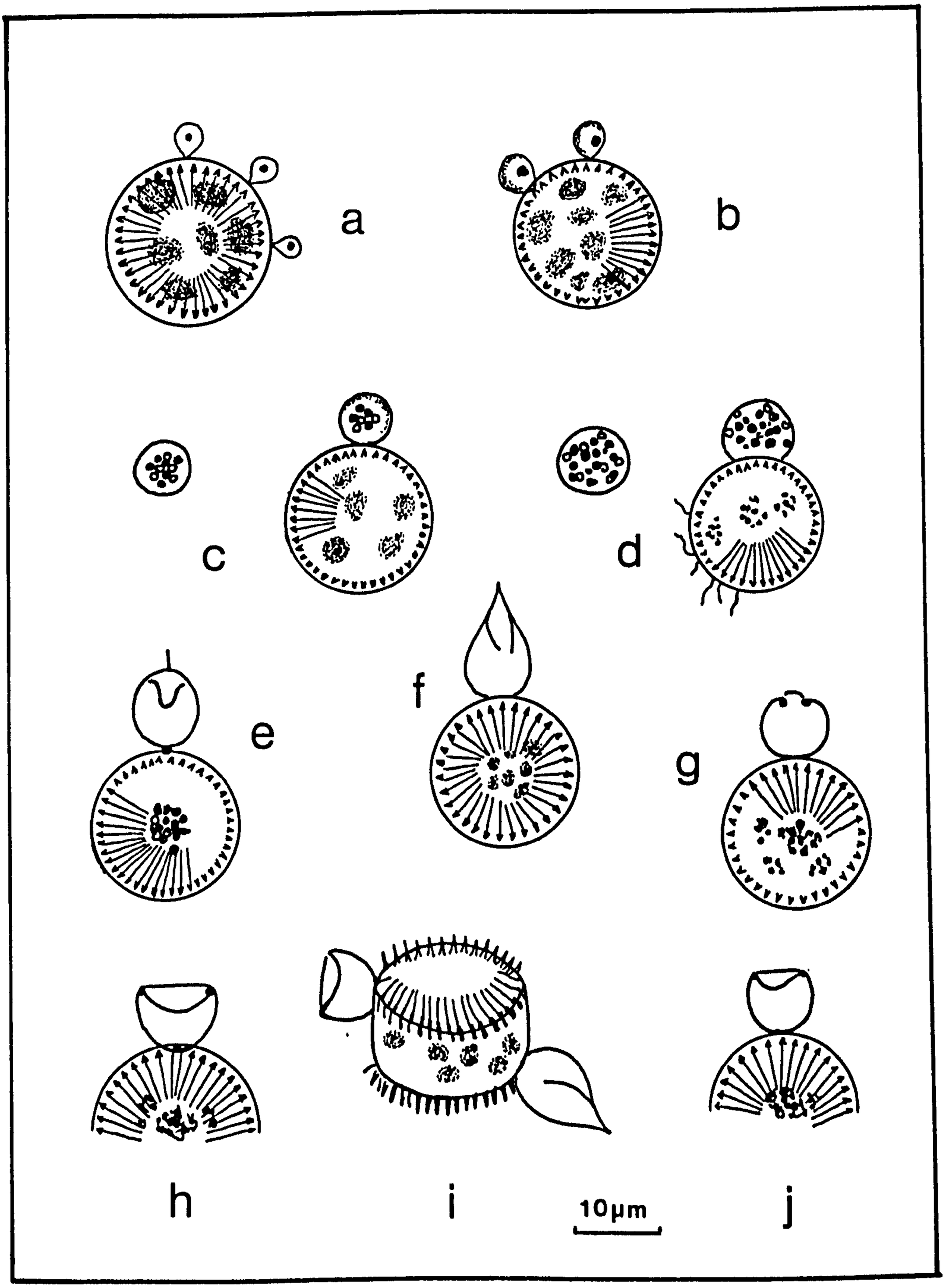
Fig.44. Fungal infection of Stephanodiscus hantzchii

a - b encysted zoospores

c - d mature sporangia

e - j empty sporangia

all drawings at X450


a
b
c
d
e
f
g
h
i
10µm
j

ends of the sphere were characterized with two thick dots. A lid-like process was present on this open part of the sporangium.

Identification of the fungus was quite difficult since little data is available on its full development. However, the mature sporangia resembles that of the fungus found on Cyclotella spp. in this study. S. hantzchii and Cyclotella spp. were most frequently present in the same time period. Nevertheless it is impossible at this stage to determine whether they are the same fungus or not without more data on the full life cycles of both fungi.

SPARROW (1951) has described a severe epidemic of Podochytrium cornutum Sparrow on Stephanodiscus niagarae Ehr., a diatom which is not recorded for Britain (CANTER & LUND,1953). He stated that up to sixty fungal thalli in varying stages of development may occur on one cell but death is caused by a single infection. An operculate chytrid which resembles Zygorhizidium planktonicum Canter was reported by CANTER & LUND (1953), parasitising Stephanodiscus astraea (Ehr.) Grun. var. minutula (Kütz) Grun. in the English Lake District. In addition CANTER (1970) also reported P. cornutum Sparrow growing on S. rotula (Kütz.) Hendey and REYNOLDS (1973) found it on a S. astraea population. Fungal parasitism of Stephanodiscus astraea and S. hantzchii with the maximum infestation of 3% was reported by YOUNGMAN et al. (1976) from phytoplankton in Farmoor Reservoir.

Parasitism and Epidemics

Fungal infection of Cyclotella and Stephanodiscus started on healthy and actively growing host populations thus suggesting that both fungi tend to be parasitic.

Encysted zoospores were usually found on healthy cells with well developed chromotophores. A maximum of eight encysted zoospores were found on a single cell of Cyclotella. In such a cell, the chromotophores appeared to be reduced. Encysted zoospores were always accompanied by sporangia on the cells of Stephanodiscus. The cells, bearing sporangia, were most frequently dead although some still appeared to be healthy. There was also additional bacterial infection on both diatoms during the periods of fungal infections. In dead cells, remains of chromotophores gathered near the centre of the cells. Maximum of four sporangia were found on a single cell of Cyclotella and two sporangia in the case of S. hantzchii. Various development stages of both fungi may be seen to be attached to any parts of the frustules of diatoms.

Table 8. Fungal infections of Cyclotella and S. hantzchii.

Date	Infected Diatom	Infection Degree (%)
1978. 4th Sept.	Cyclotella	1.2%
19th Sept.	"	0.4%
1979.19th Feb.	"	0.3%
5th Mar.	S. hantzchii	0.3%
2nd Apr.	Cyclotella	0.2%
1980. 7th Jan.	S. hantzchii	2. %
4th Feb.	"	0.4%
13th Oct.	"	2. %
10th Nov.	"	0.9%
8th Dec.	Cyclotella	0.3%
1981. 6th Jan.	"	1.4%
19th Jan.	"	0.8%
2nd Feb.	"	1.3%

As is seen from the above table the fungal infection on both diatoms was quite low, maximum degree of infection of 1.4% and 2% were recorded on Cyclotella and S. hantzchii respectively. Most of the fungal infections were recorded in autumn and winter periods. During the periods of fungal infection, although the numbers of both diatom were quite high, growth-rate of the fungi was exceedingly slow. This suggests that rate of active growth of diatoms was far faster than those of the fungi.

Infection on S. hantzchii consisted mainly of sporangia particularly empty sporangia while more encysted zoospores were

found, besides high numbers of sporangia on Cyclotella. No effect was detected on the growth cycles of the host populations since the degree of infection was very low.

The occurrence of the parasite on Cyclotella coincided with a wide temperature range from 3 - 18°C. while the fungus on S. hantzchii showed a better relation with temperature, occurring mostly at low temperatures between 4 - 7°C (Fig. 1). When both fungi appeared on diatoms, water level was either already high or rising and pH level varying between 7.5 - 7.7 (Figs 1 & 2).

Dissolved nutrients also showed some regular features when these fungi appeared (Fig.3). The parasite of Cyclotella occurred mostly at low concentration of silica whilst nitrate and phosphate were either high or low. The occurrence of the fungus on S.hantzchii showed a clearer relation with dissolved nutrients. Concentrations of nitrate and silica were high while that of phosphate was low.

Summary and conclusions

Fungal parasites of Cyclotella spp. and Stephanodiscus hantzchii are regular in occurrence, usually coinciding with periods of active growth of host populations.

Infections are relatively low despite the high cell numbers of the hosts, suggesting a slow growth rate of the parasites.

Both parasites usually occurred under similar physico-chemical conditions.

Fungal infections are too low to exert any conspicuous effect on the growth of the host populations.

Fungal infection on the genus Melosira

The genus Melosira was represented by four species: M. ambigua, M. granulata, M. granulata var. angustissima and M.varians, in the present study. All these filamentous diatoms exclusive of M. granulata var. angustissima, were infected by fungi on a few occasions over the study period. Fig. illustrates the infection on Melosira spp. The identification of the fungi was quite difficult since only small parts of the developmental stages were observed. These algae never exceeded 1% on any occasion and the fungus was not necessarily started on growing populations. The fungal infection on Melosira ambigua was very slight although the alga was present on most occasions (Fig.45). Fungal infection of the filaments was very low indeed and consisted mainly of encysted zoospores, however in two instances a mature sporangium and a resting spore were also observed. The encysted zoospores were either elipsoid, 4 - 5μ high, 2 - 3μ wide, with a short germ tube or dome-shaped measuring 2 - 3μ high by 3 - 4μ broad. The zoospores with a single oil globule simply fix themselves on the filaments. Sessile mature sporangia are circular 10 - 11μ in diameter, enclosed by 8 - 9 zoospores, pale green in colour. Resting spore is almost spherical, 8μ in diameter, with a thick smooth wall and internally granular, containing a small globule. Infected cells still appeared to be healthy. The fungal infection of M. ambigua usually coincided

Fig.45 (a - d) Fungal infection of Melosira ambigua

a - b encysted zoospores

c mature sporangia

d resting spores

(e - i) Fungal infection of M. granulata

e encysted zoospores

f - g developing sporangia

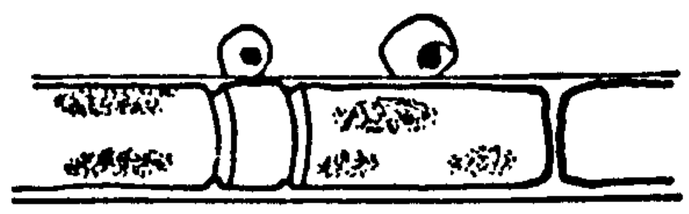
h - i mature sporangia

(j - k) Fungal infection of M. varians

j encysted zoospores

k immature sporangium

all drawings at X450



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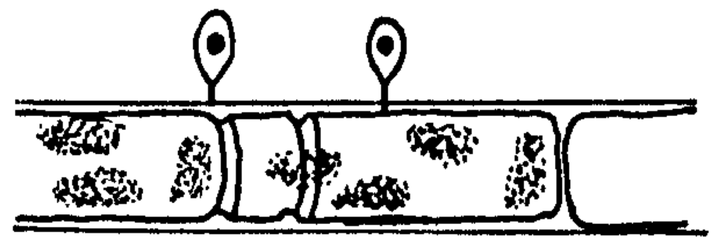

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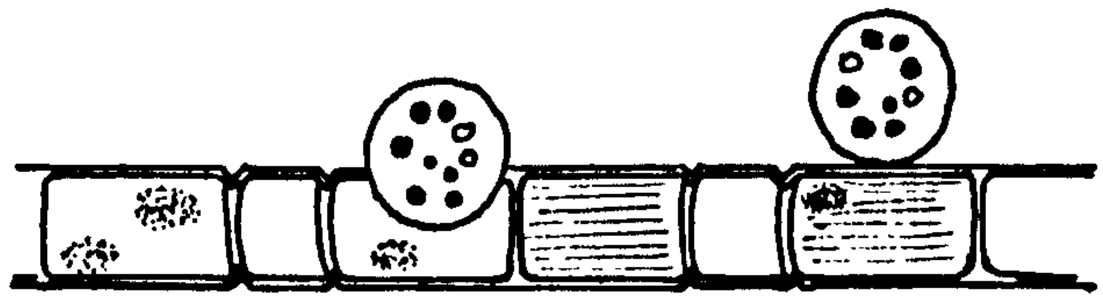

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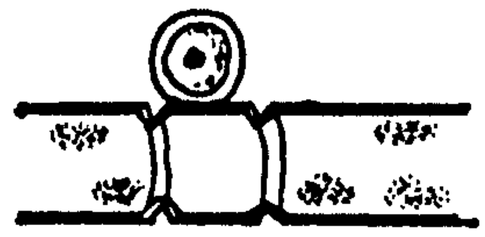

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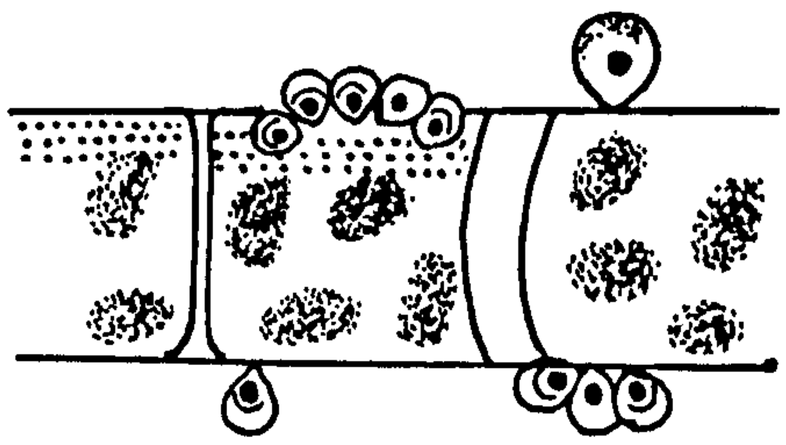

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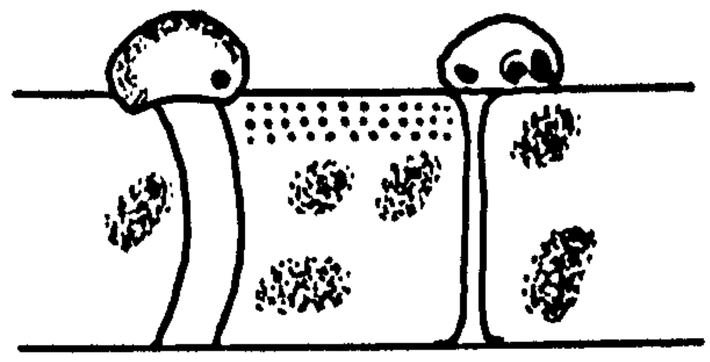

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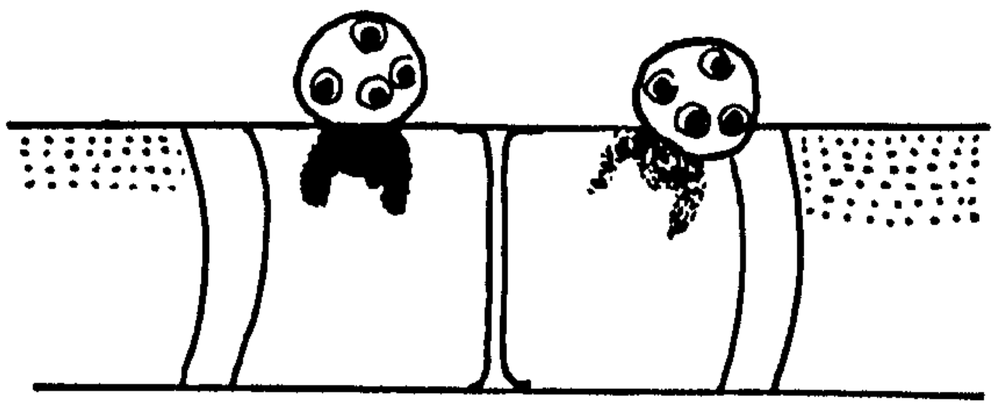

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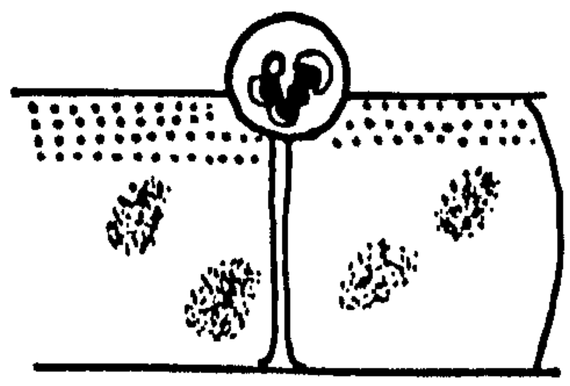

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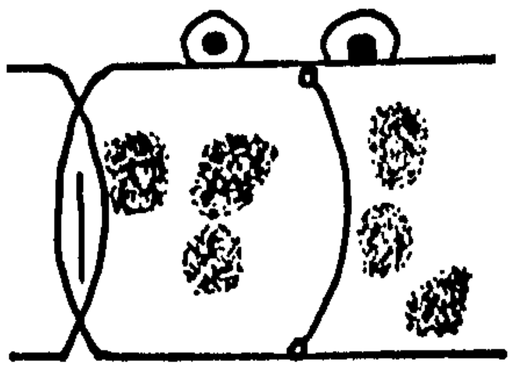

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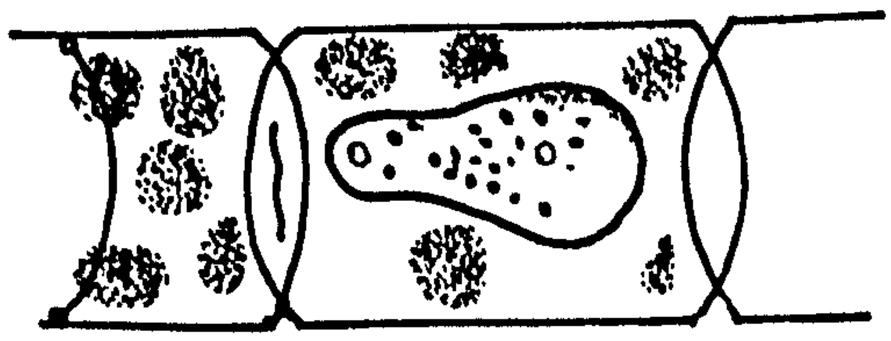

k

10 µm

with periods of parasitism of Asterionella formosa by Zygorhizidium affluens. The two fungi appear to be similar as far as the life stages could be observed, hence they might be the same fungus. Infections on M. ambigua were recorded on 12th December 1978, on 9th January, 2nd, 16th, 30th April and 26th November 1979.

Filaments of M. granulata were encountered in most samples of Shearwater generally in low numbers.

Fungal infection on the filaments was usually caused by encysted zoospores. However mature sporangia were only rarely found. The encysted zoospore (Fig. 45e) is obovate in shape, 2 - 4 high, containing a single oil globule. A nuclear cap is visible in some stages. Zoospores were observed attached to filaments without a visible germ tube. At one stage a cylindrical young sporangium was observed. Two different shaped mature sporangia were found; one is spherical, 9 - 10µ diam.,another oval 5µ high by 5µ broad, both were sessile (Fig. 45h,i). They contained 4 - 5 zoospores within the sporangium. Infected cells of M. granulata were mostly dead or with disorganised chromotophores. However dead filaments were also found without any infection throughout the year. Fungal infections were recorded on 19th September 1978, 16th April, 26th November 1979 and 6th January 1981. This fungus appears to be similar to that on Cyclotella kutzingiana (Fig. 43) whose infection at one stage coincided with the period of infection of M. granulata.

M. varians occurred in lesser numbers and in fewer samples in Shearwater compared with the occurrence of either M. ambigua or M. granulata (Fig. 16).

Only one filament of M. varians was found to be infected (on 9th January 1979) throughout this investigation. It is quite difficult to determine from the illustration whether or not these life stages belong to the same fungus. The encysted zoospore has a single oil globule and is spherical 3μ in diameter. The young sporangium is pyriform 15μ high by 4μ (apex) - 8μ (base) wide, and contains many small granules. It resembles that of the chytrid on Microcystis aeruginosa. However it is noteworthy that there was no infection on this alga during this period. The infected filament still appeared to be healthy with little disorganized chromotophores.

Infection of genus Melosira

A new chytrid Zygorhizidium melosirae Canter was described by CANTER (1950; 1967) as a parasite of Melosira italica in the English Lake District. PATERSON (1956) described Rhizophidium pedicellatum Paterson which was also parasitic on Melosira. The fungi found on Melosira spp. in this study, however, did not appear to resemble the chytrids described by those authors at least as far as data is available for the mature sporangia.

Summary and conclusions

Fungal infection on the representatives of the genus

Melosira was quite low, never exceeding 1%.

Infections occurred on growing as well as declining host popuations and consisted mainly of encysted zoospores and mature sporangia.

Parasites definitely do not follow the seasonal cycles of host diatoms and were absent during the periods of high numbers of host populations. This may well be the reason for the very slow development of the parasites since their occurrence usually coincided with low numbers of host populations.

Fungal infection of Chlorophyceae

The members of this class were outstanding in the phytoplankton of Shearwater and some of them were infected by fungi from time to time. Fungal infection of individual species is considered separately.

Fungal infection of the genus Coelastrum

The genus Coelastrum was represented by two species in Shearwater: C. reticulatum and C. microporum; which were both infected by a morphologically similar fungus from time to time. In searching through the literature I failed to find any reports of a fungus which grows parasitically or saprophytically on these colonial green algae.

Parasite of C. reticulatum (Dang.) Senn

Spherical coenobium of C. reticulatum are surrounded by a wide gelatinous sheath. The fungal thallus is epibiotic, monocentric. The sporangium develops from the encysted zoospore and possibly part of the germ tube (Fig. 46 & 47).

The encysted zoospores mostly come to rest on the gelatinous colonial envelope of the host and sends a delicate germ tube (3 - 10μ long) into the gelatinous sheath (Fig 46o). The tube grows inward until contact is made with the host-cell contents. The germ tube gradually broadens until it is almost cylindrical, slightly ovoid (Fig. 46b-e & 47b-c) and its protoplasm may contain a vacoule-like structure (Fig. 46k). When mature it varies in shape from pyriform (Fig. 46j,l) measuring 3 - 10μ high by 5 - 6μ broad, to spherical (Fig. 46m) 8μ in diameter. The sporangium is sessile or stalked on the cell or colonial envelope (Fig. 46, j-m & 47d-f.). If there is a stalk it may possibly be the part of the original germ tube (Fig. 46o). The sporangium may contain from 8 to 20 zoospores which are either closely or distantly arranged. The zoospores are fully formed within the sporangium. The dehiscence was not observed. However there is strong indication that it may be apical considering the spherical sporangium and the apex of the empty sporangium. Empty sporangia appeared as if the top part was detached as a lid (Fig. 46n). The sporangium does not collapse after the dehiscence but a few faults may occur in the sporangial wall (Fig. 46o).

Resting spore formation was observed when an occasional trip

Fig.46. Fungal infection of Coelastrum reticulatum

a encysted zoospores

b - d developing sporangia

g - h immature sporangia

j - m mature sporangia

n - o empty sporangia

p resting spore formation (m = male)

(f = female)

all drawings at X450

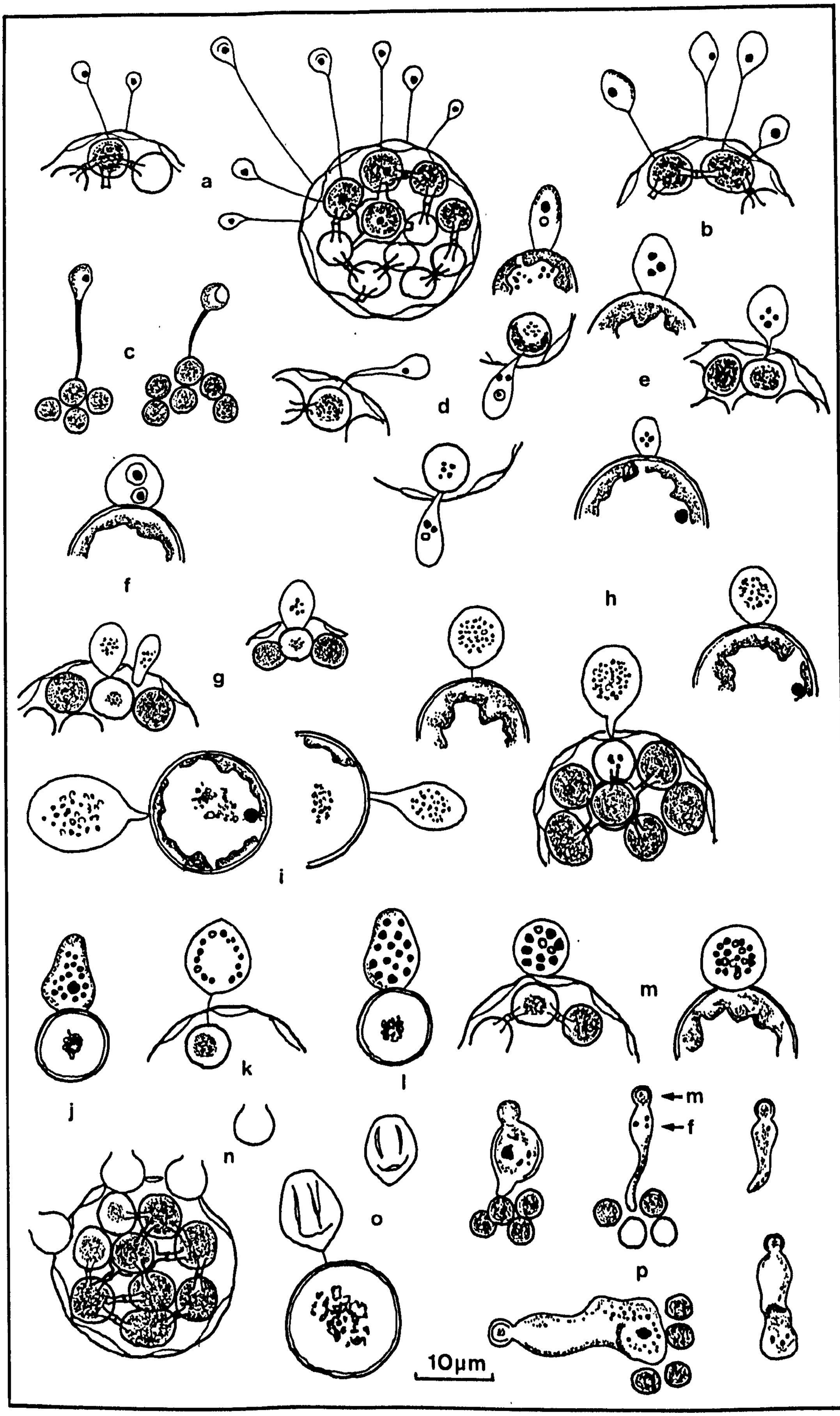
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Fig.47. Micrographs of fungal infection of C. reticulatum

a - c developing sporangia

d - f mature sporangia

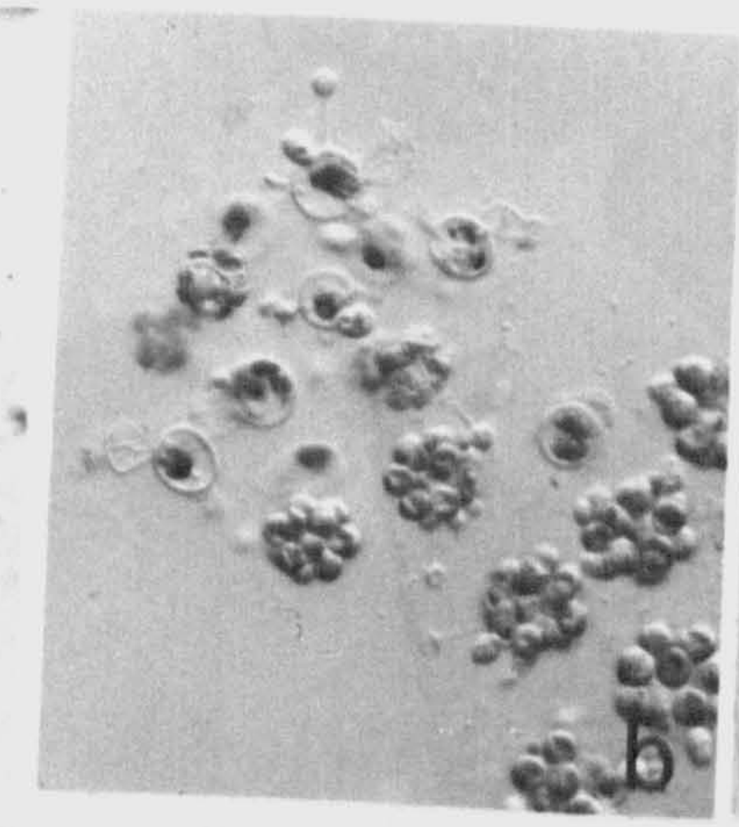
g - i fungal infection of C. microporum

j - k resting spore formation on C. reticulatum

a - j at X450

k at X860


a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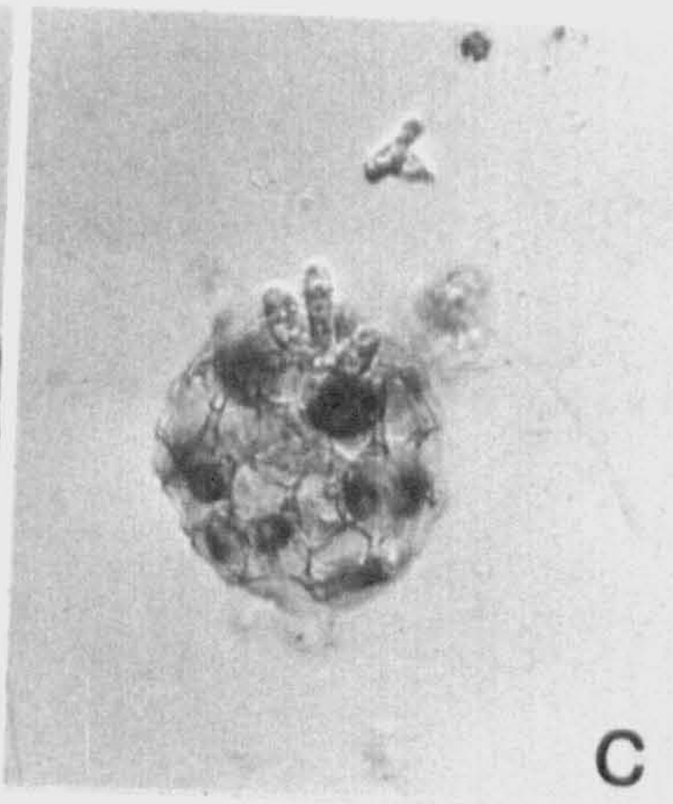
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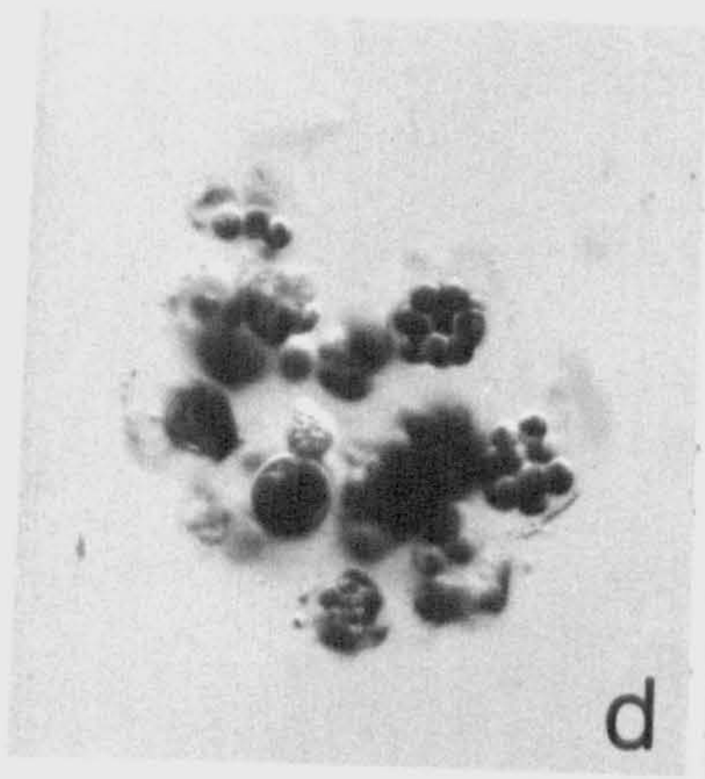
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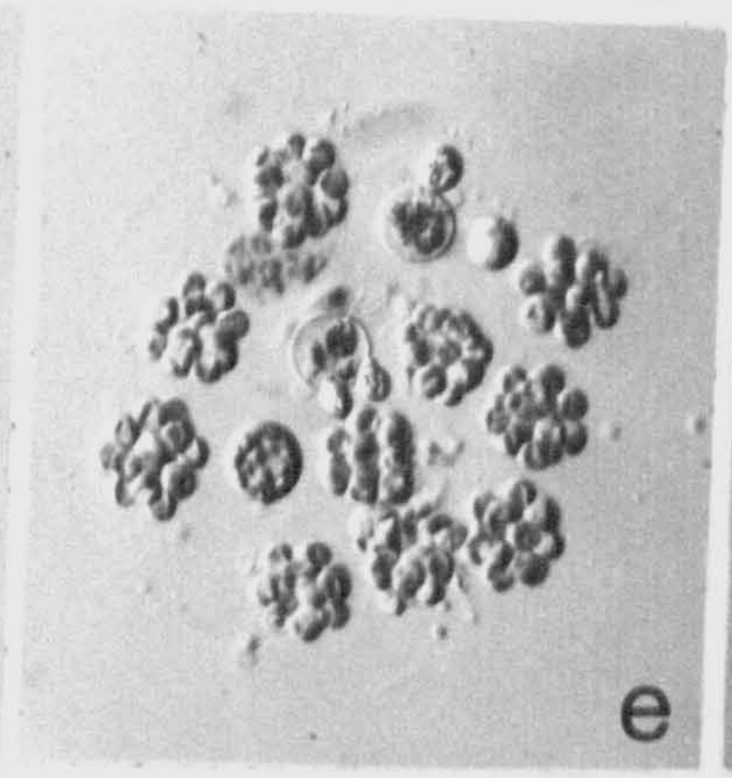
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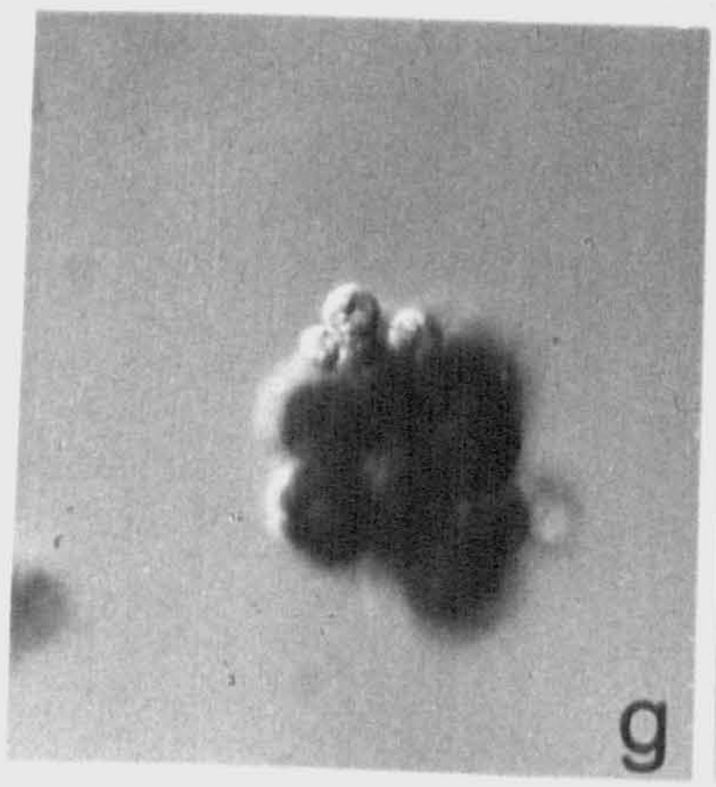
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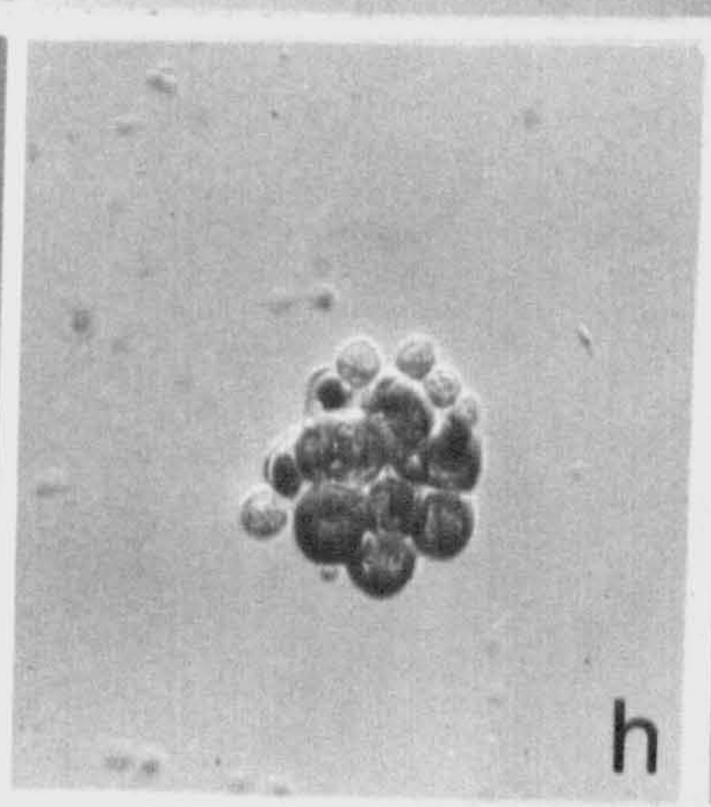
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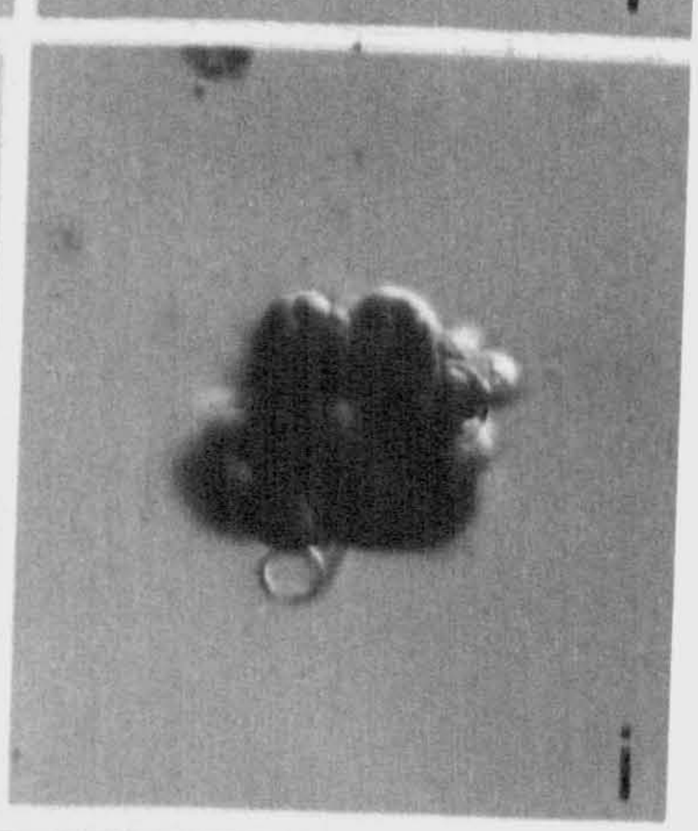

i

j

k

was made to Shearwater in order to obtain more data on fungal parasitism of algae after the present investigation ceased. It was thought to be worth-while to include this in this section.

Individual swimming gametes were not observed. However a male gamete can be seen attached to the receptive female gametengium (Fig.46p). The flagellum of the male gamete was probably withdrawn. In some cases the male thallus was observed to be fused into the female thallus and appeared as a blob or small spherical body (Fig 47j,k). This suggests that the fungus was probably undergoing a sexual reproduction. Resting spore formation appeared to be similar to that which is observed in the chytrid Chytridium (see WEBSTER, 1970, pp.45.) where the resting spore is endobiotic. The actual resting spore of the parasite of C. reticulatum was not observed.

Parasite of C. microporum Näegeli

The spherical coenobium of the alga is composed of sheathed globular or sometimes ovoid cells. The zoospore with a large oil globule encysts on the cell (Fig.48a,b) and the nuclear cap is usually visible (Fig.48a,c). A few stages with encysted zoospores having a fine germ tube were observed (Fig.48c) although they are mostly seen without tubes. The sporangium is formed by direct enlargement of the zoospore. The young sporangium varies in shape as it may be dome-shaped (Fig.48g), ovoid (Fig.48i) or pyriform (Fig.48f). In a few cases vacuole-like process were also observed in the protoplasm (Fig.48f). The mature sporangium is either ovoid (Fig. 48k)5μ to 10μ high, 5μ to 7.6μ broad or spherical

Fig.48. Fungal infection of Coelastrum microporum

a - b encysted zoospore

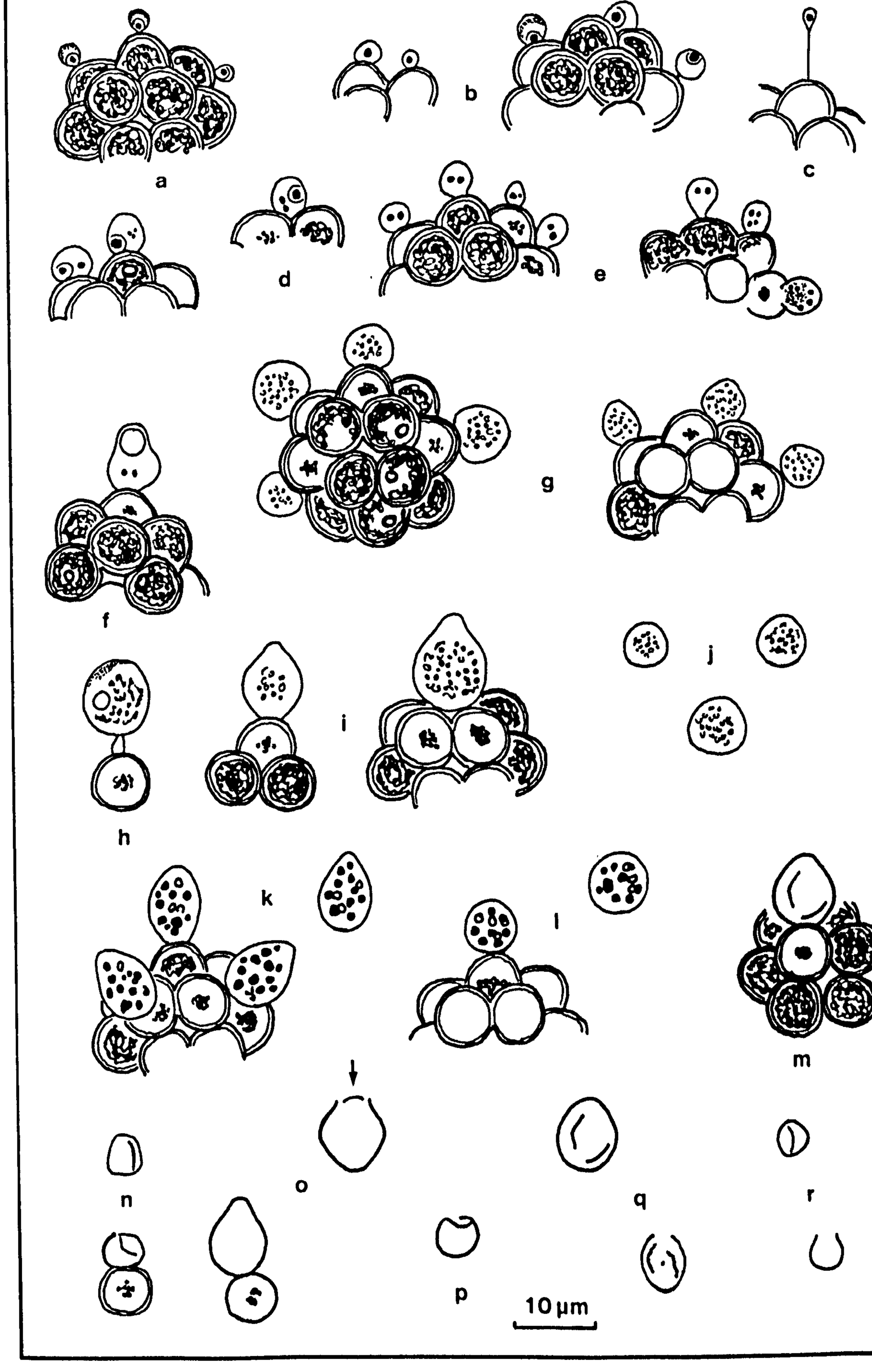
c - f germinating zoospore

g - j immature sporangia

k - l mature sporangia

o - r empty sporangia

all photographs at X450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(Fig.481) 8μ in diameter. There may be 7 - 20 oil globules each marking the position of a zoospore (Fig.481). The sporangium is sessile on the cell. Rhizoids were not observed even in dead cells whose contents were very much reduced to a few small greenish spheres (see dead cells). If there is a stalk as was observed at a few stages when the sporangium was young (Fig.48h) this might be the part of the germ tube of the encysted zoospore which was also rarely observed (Fig.48c).

Dehiscence has not yet been established; however, the evidence so far suggests that the unthickened apex of the sporangium probably separates as a lid. A lid on the apex of an empty sporangium was occasionally found (Fig.48o). This suggests that the fungus is probably operculate. The sporangium remains intact after the dehiscence and a few faults may be seen occasionally on the sporangial wall (Fig.48 m-r).

Resting spores were not observed.

Fungal thalli, growing on both Coelastrum reticulatum and C. microporum appear to be very much alike in their life cycles. The encysted zoospores of both fungi more or less displayed the same features. The germ tubes of the zoospores seemed to be the only distinguishing difference between the two fungi. However, this may be due to the fact that one has to penetrate the gelatinous colonial envelope (C. reticulatum) in order to reach the cell whereas the other one just settles on the coenobium of C. microporum which lacks such an envelope. Nevertheless a zoospore with a fine germ tube was also observed on the latter alga. The sporangia of both fungi also showed the same characteristic features; both develop from encysted zoospores, sessile or stalked, ovoid or spherical in shape. After dehiscence

both sporangia are still rigid and display the same kinds of faults on the sporangial walls. Considering all these similar features of the fungi, one might suggest that they are the same fungus. In addition, both fungi occurred in the same period on these very closely related algae. Further observations such as dehiscence, individual zoospores and resting spore formation, are needed to find out whether they are truly the same fungus or merely close relatives.

Parasitism and epidemics

Both fungi, growing on Coelastrum reticulatum and C. microporum showed the characteristic features of parasitism. The zoospores always attacked the healthy coenobia of both algae. At length, by gradual development of the sporangia, the fatal consequence of parasitism occurred on the infected cells. This suggests that both fungi are parasitic in nature.

Development phases of both fungi in relation to epidemics are shown in table 9.

	Coelastrum reticulatum									C.microporum		
	1978				1979				1980	1979	1980	
Dates	30.4.	30.5.	11.6.	12.12.	9.1.	29.10.	12.11.	26.11.	29.9.	17.9.	29.9.	13.10.
Total Coe.counted	50	50	50	50	50	50	120	130	60	50	60	50
% infection	4	4	6	2	2	4	26	36	13	2	13	28
Encys. zoospores	1						37	37	3	1	5	8
Germ. "	2	1	1		1	1	11	8	1		5	7
Imm. spo.		3	7	1				12	3		12	9
Mat.sporan.			1			1	4	3	4	3	1	3
Empty sporan.								30	4		8	7

Table 9 Developmental phases of parasites of C. reticulatum and C. microporum during their occurrence in Shearwater.

It is apparent from the above table that the parasite of C. reticulatum was still at an early stage of development whenever it occurred in small numbers. Highest numbers of sporangia were recorded during the most severe epidemic (1979). During the infection of C. microporum there was not an outstanding increase in the numbers of sporangia, and developing sporangium were higher in numbers. The cells infected by encysted zoospores were still healthy in appearance while the ones bearing sporangium were generally dead with small granular remains of cell contents (see dead cells).

The occurrence of C. reticulatum was far greater than that of C. microporum in Shearwater, both usually occurring in small numbers (Fig.22). Two important peaks of the former were interesting over three years of observation.

Occurrence of fungal parasites of both C. reticulatum and C. microporum in relation to physical factors and fluctuations in the numbers of coenobia is shown in a joint graph (Fig.49 It is apparent from the figure that coenobia of C. reticulatum were parasitized more frequently than those of C. microporum. This might appear to be due to higher numbers of the former (Fig.22). The parasite of C. reticulatum was sporadic in occurrence. Infection on this alga remained low apart from the epidemic recorded in November 1979. During this epidemic, the highest infection (36%) on C. reticulatum was recorded. The epidemic lasted nearly a month and ended suddenly after the maximum infection.

In 1980, both algae were parasitzed at the same time (29th September). Degree of infection was also the same (13%)

at the start. Infection on C. reticulatum disappeared in the next sample while infection on C. microporum reached its maximum (28%). Infection of the latter lasted only two weeks and ended sharply like that of C. reticulatum in 1979 after the maximum infection was recorded. C. microporum also succumbed to an attack by the fungus on 17th September 1979 indicating that the fungus tends to be regular in occurrence although the infection was almost neglibible (2%). Low numbers of host alga may be one of the reasons for this low infection.

Parasitism by these two fungi appeared to have no marked effect on the populations of C. reticulatum and C. microporum since the hosts increased or decreased in numbers whether the parasites were present or not.

In 1978, the sharp decrease in the numbers of coenobia of C. reticulatum may not be attributed to the low fungal infection. However this kind of sharp decrease was also recorded in 1980 without the presence of fungus on the alga (Fig. 49). It is apparent from the figure 49 that the numbers of C. reticulatum were increasing before the infection started and this was followed first by decrease then by an increase during the epidemic.

In the case of C. microporum a decrease was recorded in the numbers of coenobia during the short epidemic in 1980. However the appearance of this alga was always for a short time in Shearwater (Fig. 22).

However, an increase in the numbers of dead cells of both algae due to parasitism was apparent. The cells bearing sporangia were mostly dead during the severe epidemics. This therefore indicates that rate of dead cells rises during

Fig.49. Fungal infection of Coelastrum reticulatum and C. microporum in relation to physical factors.

□ % fungal infection of C. reticulatum

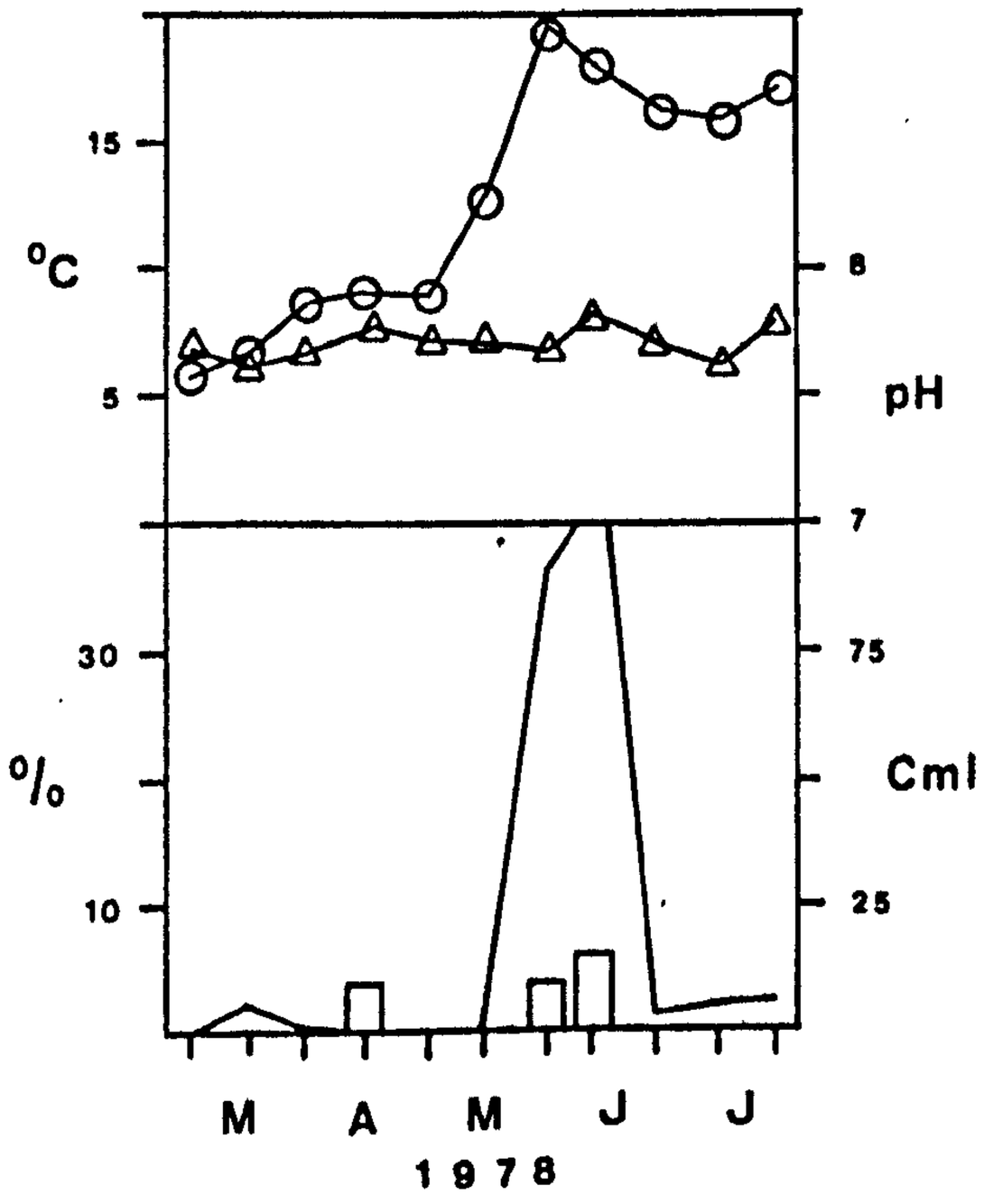
■ % " " C. microporum

△——△ pH

⊖——⊖ temperature

——— (cml) Number of Coenobia/ml of C. reticulatum

- - - - (cml) " " " C. microporum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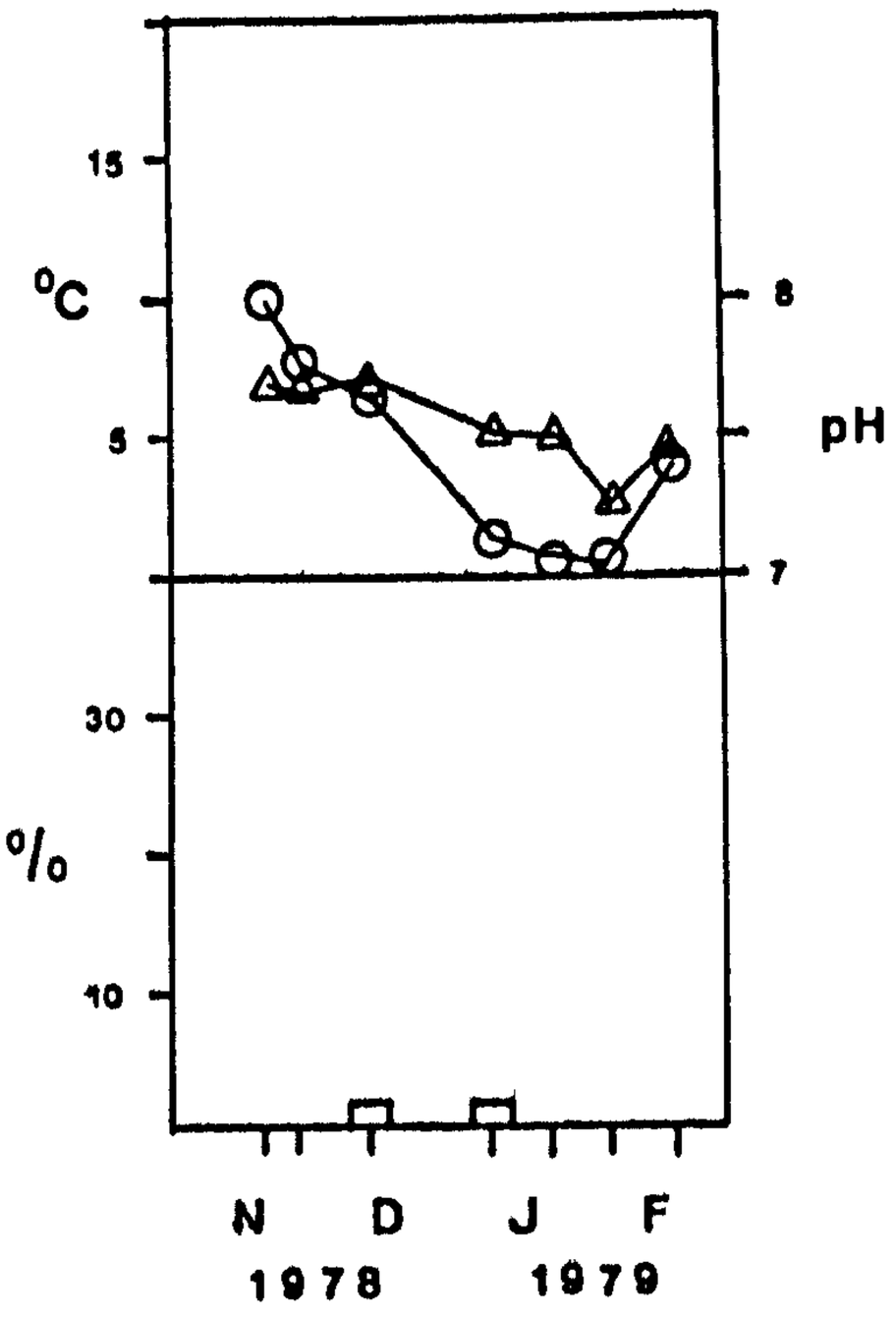
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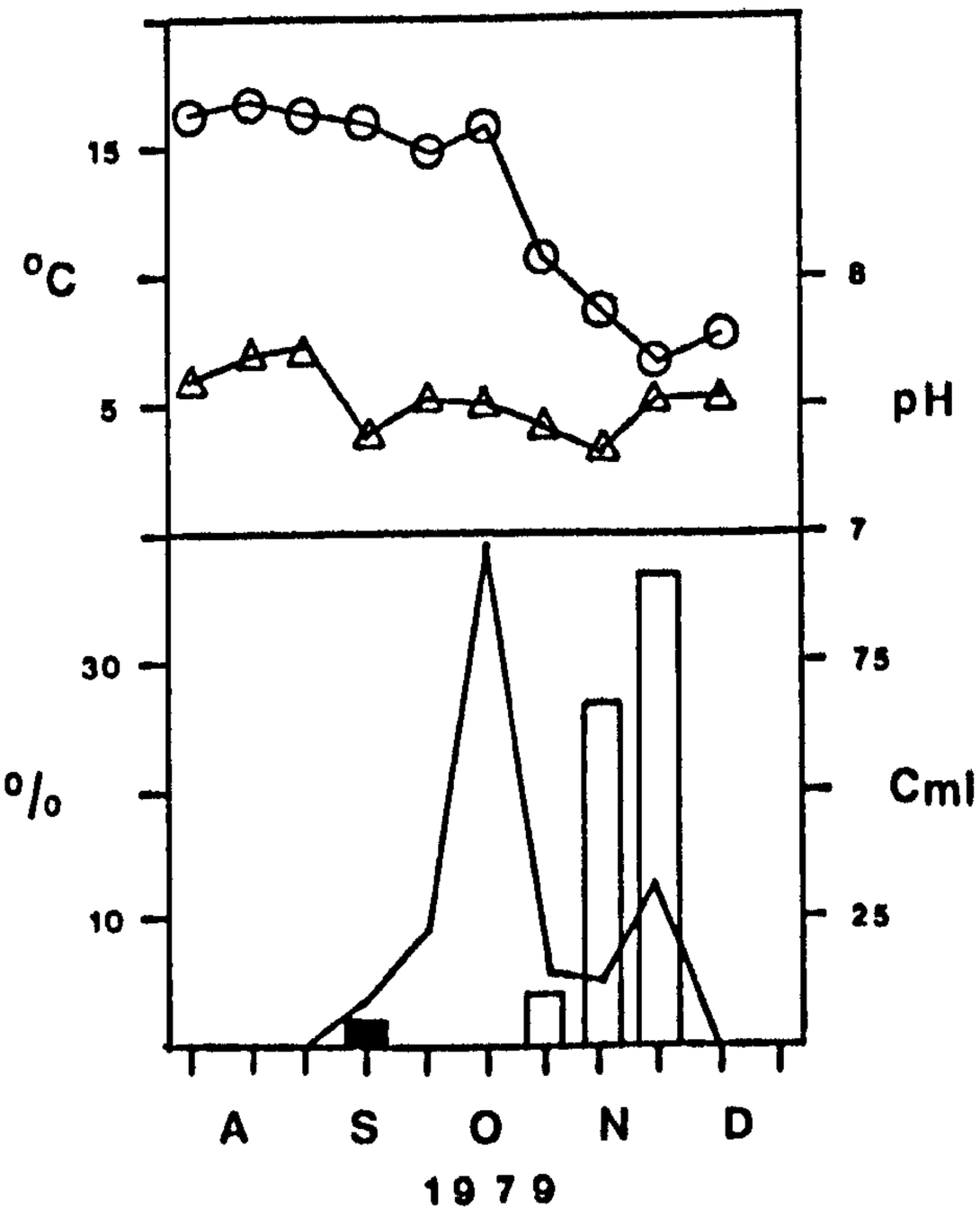
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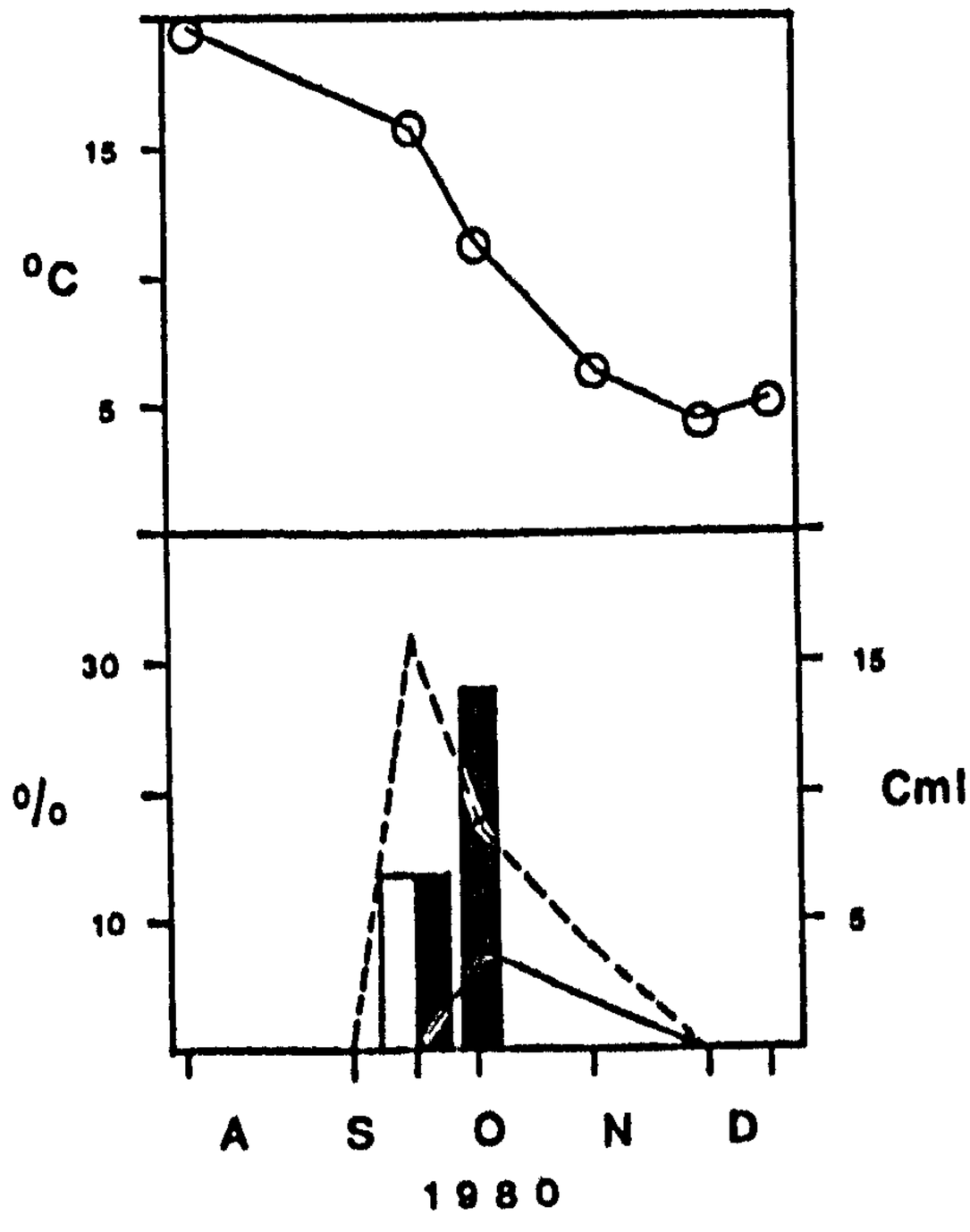
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epidemics, depending on the degree and developmental phases of the infection.

Occurrence of parasites

Occurrence of both fungi did not show a marked relation with pH and temperature in Shearwater. Their appearance was synchronized with temperature changes. But variation in pH and temperature did not appear to be important for either fungus (Fig. 49). Concerning the main epidemics, declining water temperature and rising water level seemed to be in the favour of both fungi (Fig. 2 and 49).

Dissolved nutrients, however, showed a clear relationship with the occurrence of both fungi in Shearwater. Phosphate level was always decreasing while nitrate was increasing and silica was at high levels (Fig. 3). This suggests that nitrate and silica may appear to be involved in the growth of both fungal thalli.

Identification of both fungi has not yet been established. Considering they are different species than those recorded on other algae in Shearwater, they showed a clear host specificity. Many diatoms and green algae were also present without any infections on them when C. reticulatum and C. microporum were parasitized. Pediastrum boryanum,which is also a coenobial type of green alga, was present and in far higher numbers but there was no infection on this alga (Fig.25).

Summary and conclusions

Coelastrum reticulatum and C. microporum appeared to be parasitized by very similar fungal thalli or maybe by the same fungus.

Parasites of C. reticulatum were irregular in occurrence whilst those of C. microporum occurred in the same periods. Occurrence of both parasites was synchronous with growing and healthy host populations and showed no marked relation with physical factors but rather with dissolved nutrients.

Coenobia of C. reticulatum succumbed to fungal attacks more frequently than those of C. microporum. The more abundant and more frequent occurrence of the former alga could be the cause for this. Both fungal parasites showed a clear host specificity.

Maximum infections of 36% and 28% were recorded in the case of parasitism of C. reticulatum and C. microporum respectively .

Highest numbers of sporangia coincided with the most severe epidemics.

Parasitism showed no marked effect on the growth of host algal populations since the hosts increased or decreased in numbers regardless of the fungal infection.

Increase in the numbers of dead algal cells was apparent during parasitism.

Fungal infection on Dictyosphaerium pulchellum Wood

D. pulchellum was not a common member in the phytoplankton

of Shearwater, only becoming conspicuous in spring (Fig. 23).

Fungal infection was recorded only once on the colonies of the alga during this investigation when 12% of the cells were infected on 21st April 1980. The infection was mainly by means of encysted zoospores although a few small sporangia were also observed. Various stages of infection are illustrated in Fig.50. The ovoid zoospore measuring 1µ to 4µ high, resembles that of the chytrid Rozella parvum. However it is worth mentioning that neither Kirchneriella obesa (the host) nor its parasite R. parvum was present on 21st April 1980. The encysted zoospore with a single oil globule sends out a fine germ tube which penetrates the wide mucilageous colonial envelope which is not always visible and finally it reaches the cell. The germ tube varies in length from 1.5µ to 7µ. Mature sporangia are either ovoid 4µ to 11µ high by 2.5µ to 6µ wide or cylindrical 12.4µ high by 3µ broad. The sporangia are sessile or stalked and contain 3 to 24 zoospores.

Few cells were infected and those observed were all dead with protoplasm which appeared to be granular. The cells bearing encysted zoospores seemed still to be healthy, green in colour but a few dead cells were also observed. A maximum of two fungal cells were found on any single Dictyosphaerium cell.

This fungus tends to be parasitic like R. parvum since the fungus attacked only healthy and growing hosts.

D. pulchellum had achieved its highest numbers (26 col./ml) for 1980 when the colonies were infected. Both the alga and the fungus disappeared after a fortnight. However such sudden disappearance of the alga was also characteristic in 1978 without any infection (Fig.23). There was a negligible increase in the

Fig.50 (a-b) Fungal infection of Oocystis lacustris

a - b encysted zoospores

(c-g) Fungal infection of Dictyosphaerium pullchellum.

c encysted zoospores

d germinating zoospores

e - g mature sporangia

All pictures at X450

a

b

c

d

e

f

g

10 µm

number of dead cells which probably results from the fact that the encysted zoospores did not develop into a large number of sporangia.

In fact these two fungi on Dictyosphaerium and Kirchneriella appeared to be very much alike concerning the development stages which were observed. The encysted zoospores and sporangia of both fungi showed the same characteristic features which are mentioned separately. Therefore it might be possible that D. pulchellum was parasitized by R. parvum or a fungus which is very alike to it.

The occurrence of this fungus coincided with rising water level. The temperature was 12.5°C and pH 7.7 (Fig 1 &2).The concentration of phosphate was low while nitrate and silica were at high levels (Fig.3).

Summary and conclusions

Dictyosphaerium pulchellum was parasitized by a chytrid which appeared to be very much like Rozella parvum.

The parasite appeared only once and only an early stage of its development occurred. It is not known why this did not develop into a full epidemic.

No marked effect of the parasitism on the growth of the host population was observed.

Parasitism of Kirchneriella obesa (W. West) Schmidle

Over the period studied, the green alga Kirchneriella obesa

was recorded only twice in the summer phytoplankton of Shearwater and on both occasions the colonies were parasitized by a chytridiaceous fungus Zygorhizidium parvum Canter. The fungus was first described by CANTER (1950) as a parasite on the colonies of Sphaerocystis schroeteri and Kirchneriella obesa in the English Lake District.

Zygorhizidium parvum Canter

Thallus monocentric, eucarpic, epibiotic and operculate. The sporangium is developed from the body of an encysted zoospore and the whole or a part of the original germ tube. The zoospore with a single oil globule makes contact with the cell by means of a fine germ tube which penetrates the wide mucilageous envelope of the alga (Fig.51a,b). Germ tube varies in length from 2 to 8μ probably depending on the distance of the zoospore to the cell. However in some cases the zoospores were also observed to have settled directly on the cells without germ tubes (Fig.51a). Finally the germ tube broadens until it is almost cylindrical (Fig.51c-f). Enlargement continues until the sporangium is formed. The sporangium varies in shape, mostly ovoid (Fig.51h,i) 3 - 8μ wide by 8 - 13μ long, occasionally pyriform measuring 7 - 13μ high, 5 - 8μ broad or cylindrical (Fig.51j,k)8μ high by 3.5μ broad. The sporangium may be sessile or stalked. If there is a stalk it is simply part of the original germ tube which has not swollen (Fig.51k). Mother and daughter algal cells were both parasitized, the former bearing the larger sporangium. Neither dehiscence nor resting spores were observed. However the presence of a lid on empty sporangia (Fig.511)

Fig.51. Developmental phases of Rozella parvum Canter on Kirchneriella obesa.

a - b encysted zoospores

c - f developing sporangia

g - k mature sporangia

l empty

(→) indicates the apical lid

all pictures at X450

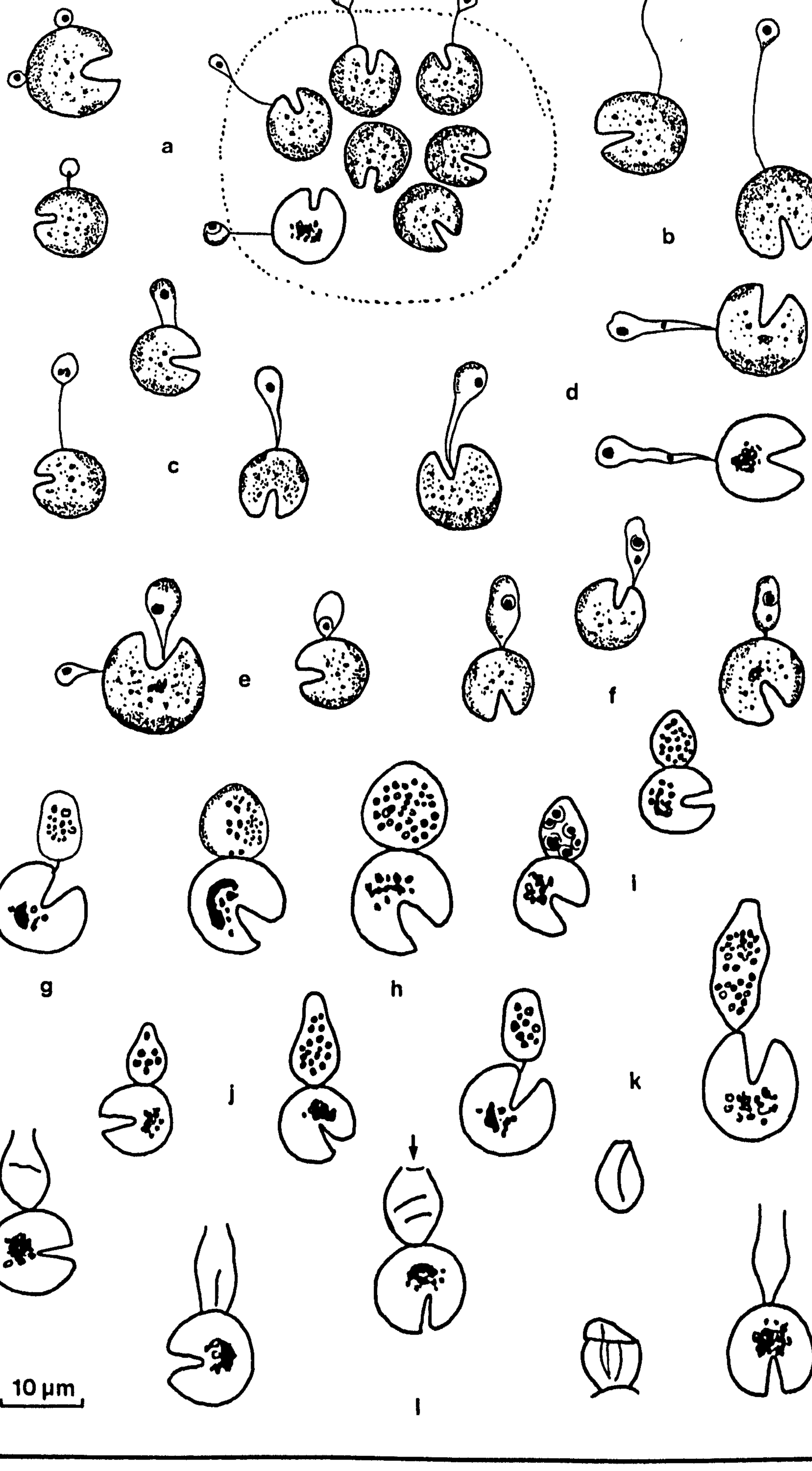

a
b
c
d
e
f
g
h
i
j
k
l
10 µm

Fig.52. Micrographs of Rozella parvum Canter on Kirchneriella obesa.

a - d germinating zoospores

e general appearance of infection

f - g developing sporangia

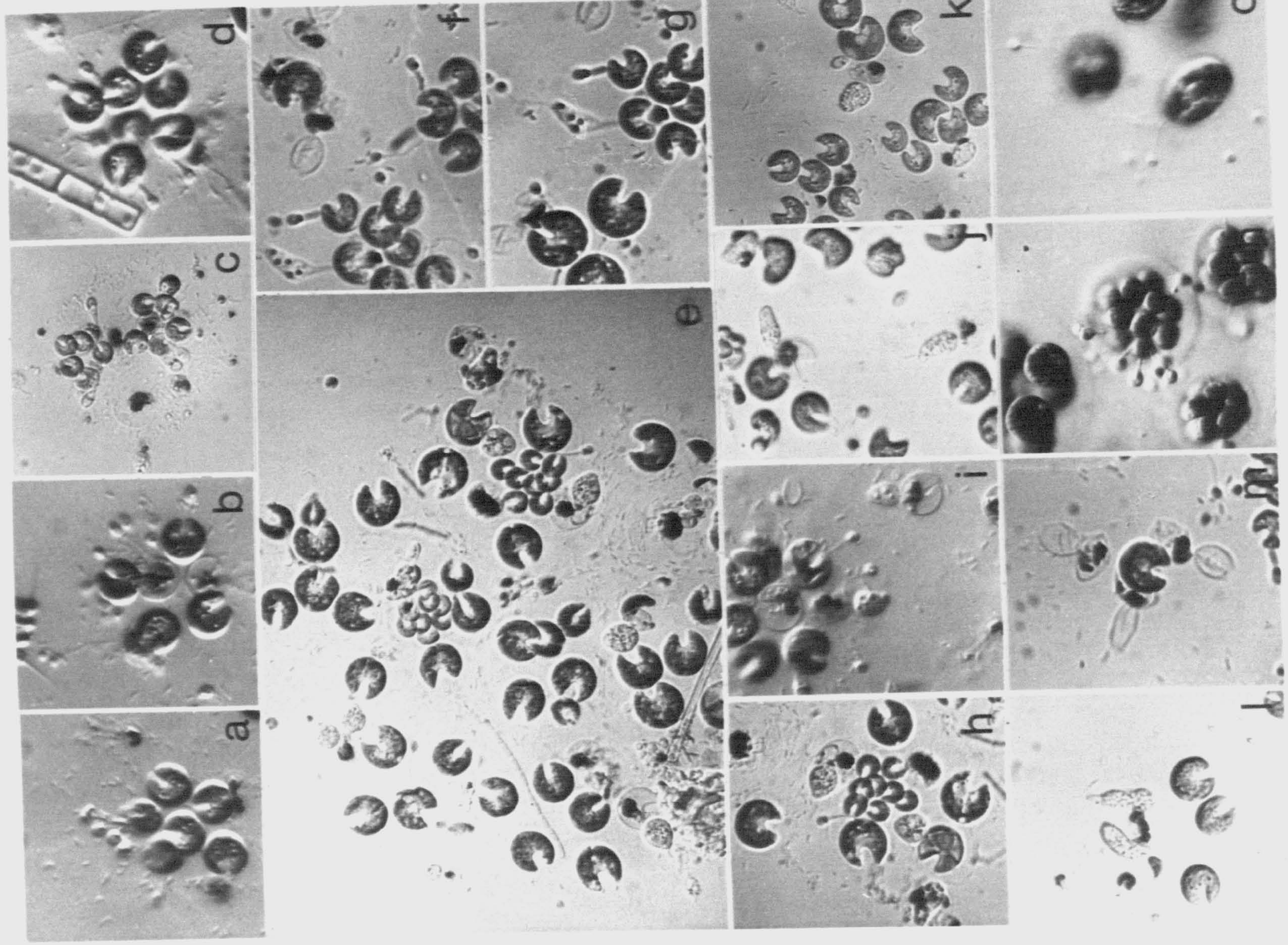
h - l mature sporangia

m empty sporangia

n encysted zoospores with germ tubes on Pandorina morum.

o encysted zoospores with germ tubes on Oocystis lacustris.

all pictures at X450


a
b
c
d
e
f
g
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j
k
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m
n
o

suggests that dehiscence may be apical by the detachment of a lid. The sporangium does not collapse after the dehiscence, however a few faults may occur in the sporangial wall (Fig.52m).

Epidemics

Development of the thallus on healthy and growing populations of K. obesa clearly indicates the parasitic nature of R. parvum.

Fig.53 shows the details of two epidemics in relation to physical factors. R. parvum tend to be regular in occurrence since both epidemics were recorded almost in the same period, June - August, in two years. Epidemics lasted only two weeks on both occasions and the host and its parasite seemed to have disappeared after a fortnight. Epidemic in 1978 was more severe (maximum 26%) than the one recorded in 1980 (maximum 8%). The highest numbers of development stages of the fungus coincided with the maximum stage of infection. Encysted zoospore and developing sporangium were dominant at the last stage of the epidemic in 1978, in contrast to epidemics of other algae (e.g. Asterionella, Microcystis, etc.).

The cells, bearing sporangia, were usually dead as they were exploited by the chytrid whereas cells bearing developing sporangia seemed mostly to be healthy depending on the stage of development. Dead cells lose their characteristic green colour and develop small granular remains of the cell contents, usually brownish-black in appearance (Fig.51g-i & 52e,j).

Infection caused an increase in the number of dead cells during epidemics. Data for the number of colonies of K. obesa

Fig.53. Fungal infection of Kirchneriella obesa by Rozella parvum.

A total number of cells counted

B ▨ healthy infected cells

▤ dead infected cells

C distribution of developmental phases of Z. parvum

☒ encysted zoospore

▣ developing sporangium

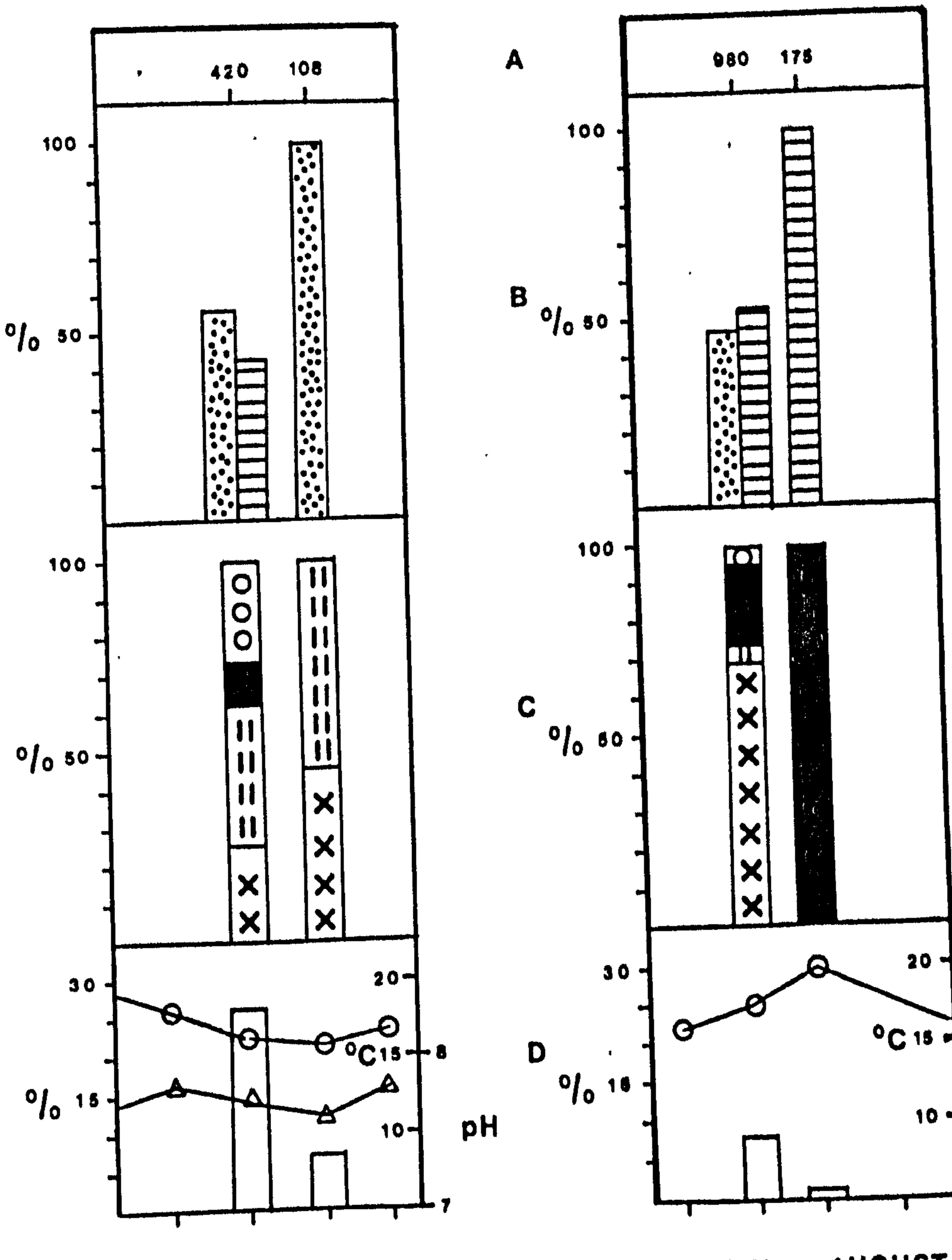
■ mature sporangium

▣ empty sporangium

D □ % fungal infection

△—△ ph

⊖—⊖ temperature


A
420
108
100
%
50
100
50
30
20
°C
15
8
%
15
10
pH
7
JUNE
JULY
1978
B
980
175
100
%
50
100
C
%
50
30
20
°C
15
D
%
10
JULY
AUGUST
1980

was available only during the second epidemic. When the epidemic started 28 col/ml were counted and there was only 5 col/ml by the end of the epidemic. The effect of parasitism on the host population remains obscure since the occurrence of the alga was always for a short time for unknown reasons.

Both epidemics occurred when the water temperature was around 16 - 20°C. This may suggest that high temperature appeared to be in the favour of R. parvum. Data for pH was available only for the first epidemic which might indicate that pH around 7.6 - 7.7 may be suitable for the growth of the fungus.

The onset of epidemics usually coincided with low concentrations of phosphate and silicate in Shearwater. Its relationship with nitrate appeared to be more complex since epidemics occurred either at high or low nitrate levels (Fig. 3) Therefore R. parvum may tend to occur even when dissolved nutrients are at low levels. Suggestions, mentioned above, also hold true for the growth of the host, K. obesa.

Summary and conclusions

The chytrid, Zygorhizidium parvum Canter, appeared to be the parasite, and its occurrence was quite regular but scarce.

Epidemics lasted only for a very short time and maximum infection recorded was 26%.

Dominant numbers of developing sporangia at late stages of the epidemics may indicate the continuation of development of Z. parvum.

Infection gave rise to an increase in the number of dead

cells.

Occurrence of the chytrid was coincident with similar physico-chemical conditions in each year.

Chytrid on Oocystis lacustris Chodat

The occurrence of Oocystis lacustris usually synchronized with spring - summer and less frequently with autumn periods in Shearwater (Fig.50).

The cells of Oocystis succumbed to slight fungal attacks on a few occasions during this study. Infection consisted only of encysted zoospores (Fig.50a,b) The zoospores appeared to resemble those of Rozella parvum, the chytrid parasitic on Kirchneriella obesa (Fig.51a), fungal parasite of Coelastrum reticulatum (Fig.46a) and of Chytridium deltanum - parasitic fungus on Oocystis spp. The zoospore cyst measures 2 - 3μ diam. and is obovate in shape. They were seen attached to the gelatinous walls of the cells or to the colonial envelope by means of delicate germ tubes which vary in length (10 - 20μ). Infection was recorded twice in June (1979 and 1980) and once in April (1978), and in September (1979). The degree of infection remained under 2% on each occasion. The number of encysted zoospores varied between 1 - 14 on single cells or colonies. Cells infected heavily were dead with granular protoplasm and circular chloroplast remains near the centre. The fungus usually attacked only healthy and growing host populations (Fig.24 &50) thus suggesting that it is most likely a parasite. Infection on O. lacustris was usually observed only

in two subsequent samples, and a fortnight later the fungus seemed to have disappeared. Factors which affected the development of zoospores into sporangia seemed to be obscure since their development stopped either at high or low level of dissolved nutrients (Fig. 3). However its appearance usually coincided with rising water level and high temperature (Fig. 1)

Parasitism and saprophytism of Oocystis spp. by fungi has been studied by a few authors. HUBER-PESTALOZZI (1944) described a new fungus Chytridium oocystidis from Switzerland growing on Oocystis lacustris. Another new Chytridium species C. deltanum was described by MASTERS (1971b) growing on several Oocystis spp. in the Delta Marsh, Manitoba. In addition, he also found three other fungi which have not been completely identified growing on Oocystis spp. In his study C. deltanum appeared to suppress host maxima and the fungus was not always able to attack rapidly growing host populations. The fungus most commonly grew on the algae at temperatures above the optimum for the alga.

Summary and conclusions

Rapid appearance and disappearance of encysted zoospores was recorded in the case of parasitism of Oocystis lacustris.

Infections remained under 2% and up to 14 encysted zoospores could be found on a single colony of the alga.

Chytrid on Staurastrum

The genus Staurastrum was represented by S. pingue and S. longipes which were encountered in the samples almost any time of the year in small numbers, usually reaching peaks in late spring and mid Autumn periods in Shearwater (Fig.27).

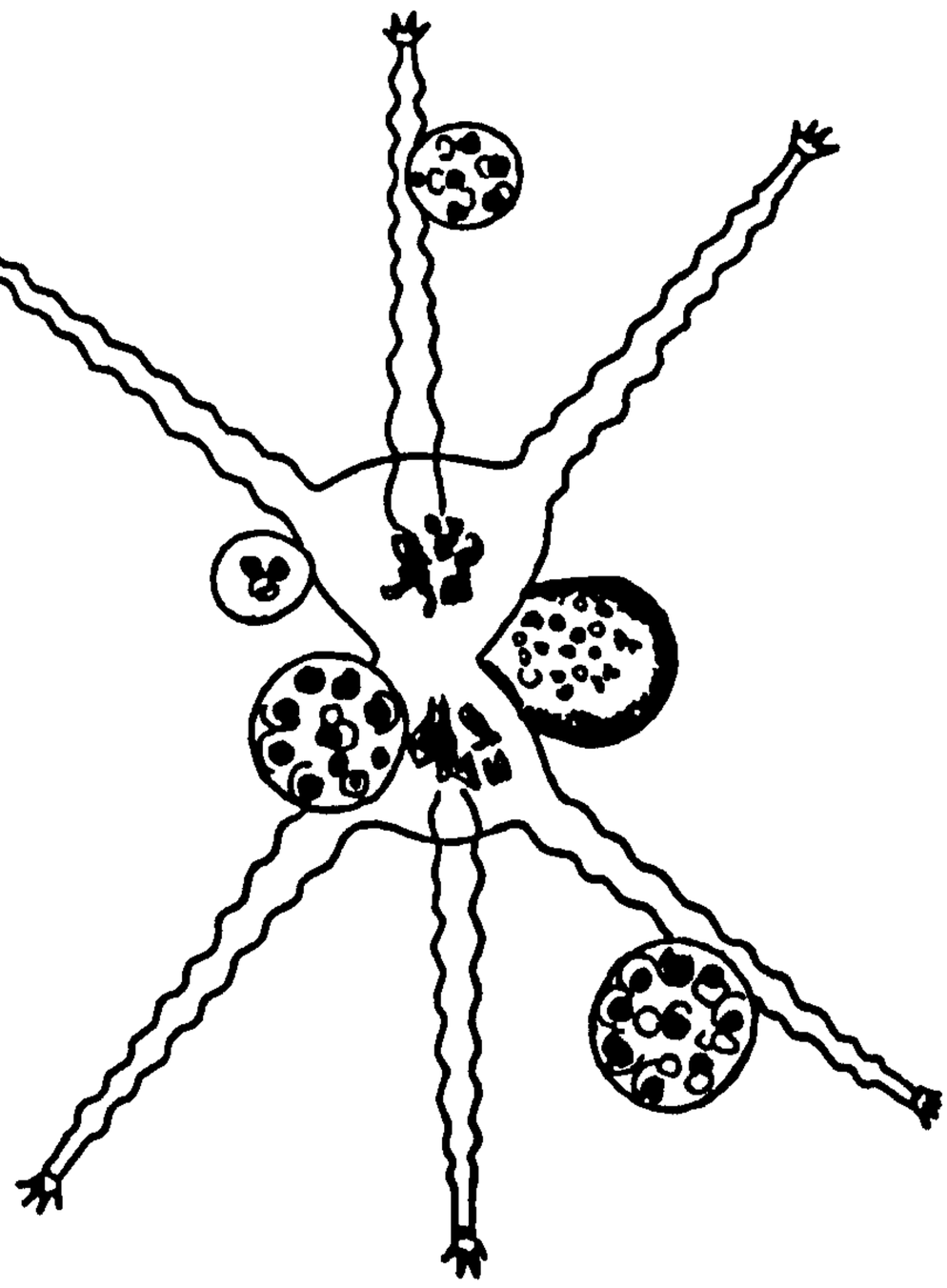
A fungus attack was observed on Staurastrum spp. only once during this investigation, when 85% of the cells were infected on 20th May 1980. Similar environmental conditions were also available for the fungus in previous years during the presence of Staurastrum, the fungus, however, was absent. The infection was quite severe and consisted mainly of mature sporangia although a few developing sporangia were also observed (Fig.54). The mature sporangium is spherical 10 - 13μ in diameter and it contains 6 - 15 zoospores. As it is seen from the illustrations, mature sporangium may occur on any part of the algae including the arms, presumably it therefore possesses a long rhizoidal system which was not observed. The infected cells were usually dead with chloroplant remains in the semicells. Often several mature sporangia were found attached to one individual. Disappearance of the fungus was as quick as its appearance since a fortnight later the fungus seemed to have disappeared. Similar incidents were also recorded by REYNOLDS (1940) for a parasite of Staurastrum paradoxum. He also found that the proportionate abundance of triradiate and biradiate facies of the desmid changed in relation to fungal infestation. He records spherical

Fig.54. Fungal infection of Staurastrum

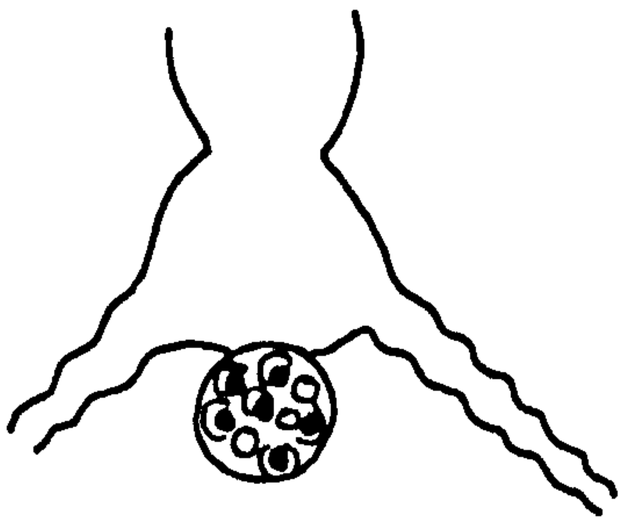
a general appearance of infection

b - c mature sporangia

all pictures at X450



a



b

c

10 µm

zoosporangia on S. paradoxum. Many parasitic and saprophytic fungi growing on desmids including Staurastrum were described by CANTER (1947, 1954, 1960 and 1961), and CANTER & LUND (1969) from the English Lake District. These fungi were most frequently biflagellate (Lagenidiales) rather than uniflagellate (Chytridiales). CANTER & LUND (1969) also studied the effect of the parasitism on the ecology of desmid populations. They found that although fungal epidemics may be severe, it does not alter the overall seasonal pattern of periodicity of the desmids but in most instances fairly dramatic declines of the algal population occur. They do concede that many species would be more abundant in lakes in the absence of such parasitism. Total disappearance of Staurastrum pingue in the Titisee following fungal parasitism was reported by SCHULLE (1970). In the present study infection was recorded when Staurastrum reached 40 cells/ml and this epidemic reduced the population to 2 cells/ml. Decrease in the algal numbers might not be entirely attributable to the fungal infection since Staurastrum also decreased sharply in the absence of fungal infection in previous years (Fig.27).

MASTER (1971e) studied the occurrence of Phlyctidium bumilleriae on growth forms of Staurastrum pingue and other Staurastrum spp. in Lake Manitoba. A three-radiate form of S. pingue, a four-radiate form and an intermediate form with three processes on one semicell and four on the other, were always found together in the summer phytoplankton. His study was interesting in that four-radiate forms were more heavily attacked by P. bumilleriae than three-radiate forms whether

the former was present in greater or smaller numbers. Intermediate cells succumbed to the fungus in proportions similar to the four-radiate form. This suggests that susceptibility of distinct growth forms of Staurastrum to fungal attack. In the present study, triradiate (3 arms on each semicell) and biradiate (2 arms on each semicell) were both present during the infection and mature sporangia were found on both growth forms. However, in the present study, it is quite difficult to determine the susceptibility of a growth form of Staurastrum to this fungus since the infection was recorded too infrequently.

Summary and conclusions

Fungal infection on Staurastrum pingue and S. longipes was encountered only on one occasion and the infection was surprisingly quite severe (85%).

Although Staurastrum spp. were present and similar physico-chemical conditions occurred in the previous years, the absence of the fungus in these years and its abrupt appearance with a high degree of infection only for one occasions remained obscure.

The fungus was already at the sporangial stage when the epidemic was recorded and caused an increase in the number of dead cells.

Chytrid on Pandorina morum (Muell.) Bory

Pandorina morum was generally a member of the summer phytoplankton in Shearwater (Fig. 20). The colonies of the alga were only slightly infected by encysted zoospores resembling those of the chytrid Rozella parvum.

Fig. 55 illustrates the infection on Pandorina morum. The encysted zoospore with a single oil globule, obovate in shape, measures 2 - 4μ diameter. It may appear from the illustrations that the zoospore comes to rest on the gelatinous colonial envelope and sends a germ tube inwards in order to make contact with the cell. The germ tube presumably broadens until the encysted zoospore is cylindrical (Fig. 55d). Immature sporangia were observed only a few times, almost spherical in shape, 8 - 10μ in diameter. Neither a rhizoid or a stalk were observed. The infection was recorded on 29th October - 12th November 1979 and on 21st July 1980 with infection rates of 4%, 7% and 8% respectively. Zoospores were dominant on the colonies which may bear 1 - 14 encysted zoospores. Infected cells still appeared to be healthy regardless of the number of zoospores; this was due to the fact that the infection was yet at an early stage.

There was also an infection on Kirchneriella obesa by Rozella parvum on 24th July 1980. These two infections seemed to be similar concerning the early stage of the development. Thus it might be possible that both algae were infected by the same chytrid Rozella parvum.

I have not found any references on fungi which grow on Pandorina morum. However the only relevant study appears to be

Fig.55. Fungal infection of Pandorina morum

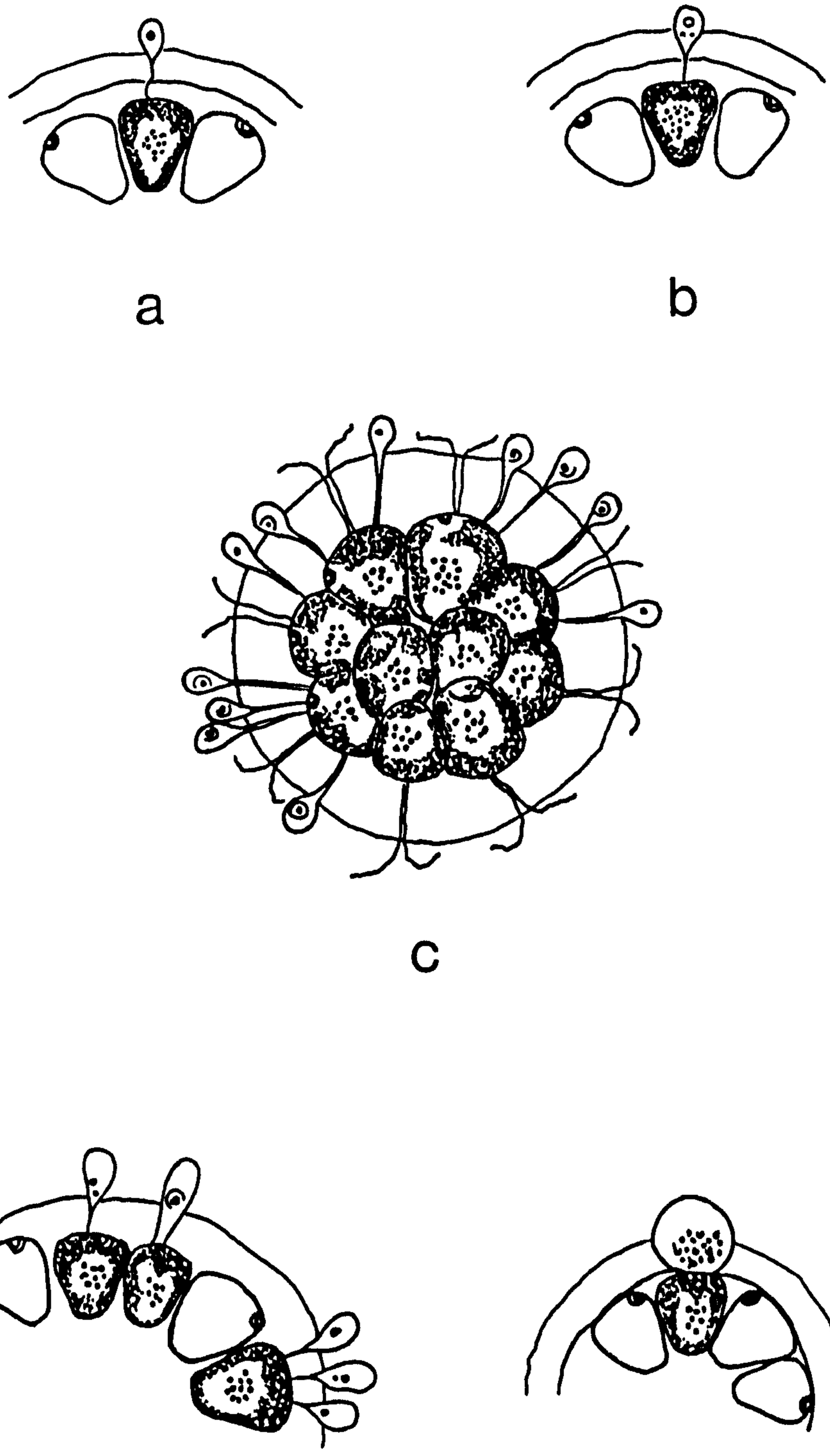
a - b germinating zoospores

c general appearance of infection

d developing sporangia

e immature sporangium

all drawings at X450


a
b
c
d
e
10µm

that of COOK (1932) who found the chytrid Rhizophydium tranversum (A.Br.) Fischer and INGOLD (1940) who described a chytrid attacking a very large proportion of Eudorina elegans, a similar type of colonial alga. These chytrids are quite different in their early development compared with that recorded in Shearwater on Pandorina morum in that they do not possess germ tubes.

Summary and conclusions

The fungus which resembles Rozella parvum was sporadic in occurrence, and its appearance was only for a very short time. Early development stages of the fungus did not continue due to unknown reasons. Maximum infection was only 8% and infections were by means of encysted zoospores.

Parasitism of Blue-Green Algae

Fungal infection on the blue-green algae was less frequent compared with the infections of diatoms and green algae in Shearwater. In fact, Microcystis aeruginosa was the only blue-green alga which succumbed to attacks of a chytrid amongst all the blue-green algae of Shearwater.

Parasitism of Microcystis aeruginosa Kuetz.; emend . Elenkin.

M. aeruginosa was a conspicuous member of the summer - autumn phytoplankton in Shearwater. During the past three

years of this study, three epidemics of a chytrid which has not yet been described, were recorded on the colonies of this alga. The same chytrid was also observed by CANTER (1954) and PATERSON (1958) on Coelosphaerium sp. (Cyanophyceae) and Microcystis sp. respectively. However they failed to give a detailed description of the fungus. In order to identify a chytrid, it is important to know all the different stages of its life cycle. Under natural conditions not all the stages can be observed at a particular moment in time. This problem can be resolved by culturing the organism. Fortunately during two epidemics I was able to observe most of the developmental phases of this chytrid.

Description of the chytrid

Thallus monocentric, epibiotic, polyphagus, eucarpic; sporangium develops by direct enlargement of the zoospore. Young sporangium is ovoid (Fig.56h) but later it becomes pear-shaped (pyriform) by the development of a tubular apical portion (Fig.56i). Its protoplasm may contain one or a couple of large vacoule-like processes (Fig.56h). The upper portion of the rudiment remains almost unchanged in width and eventually forms the neck of the sporangium which is terminated by the thicker wall (Fig.56i). The mature sporangium is pyriform in shape (Fig.56j) measuring 18 to 39μ long by 13 to 26μ broad. It may contain 13 to 28 zoospores, depending on the size of the sporangium; the ozoospores are evenly distributed in the sporangium. This is characteristic of most chytrids.

Fig.56. Fungal infection of Microcystis aeruginosa by a new chytrid species

a - f developing sporangia

g - i immature sporangia

j mature sporangia

k dehiscence (→ indicates the separation of the apex of sporangium)

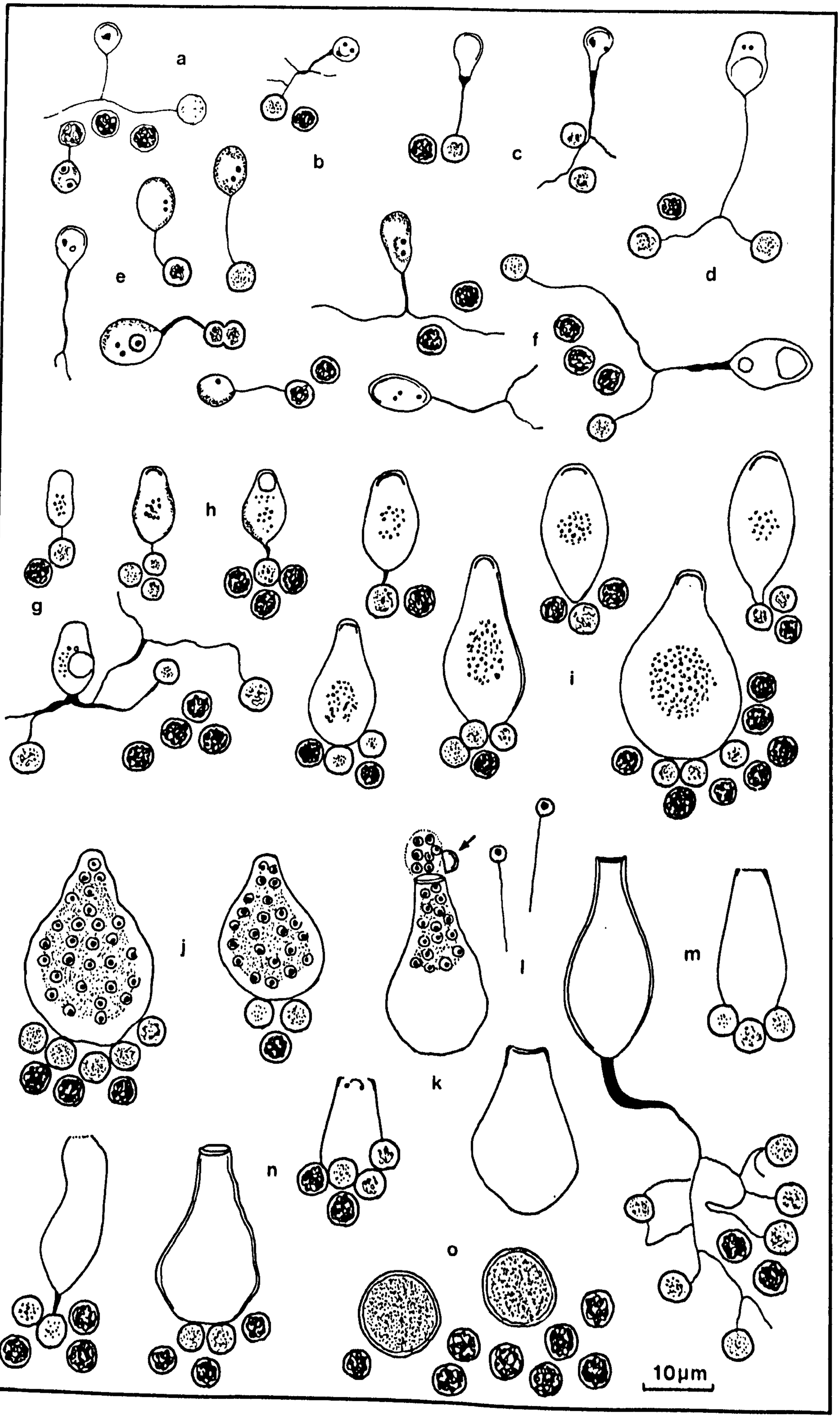
l zoospores

m - n empty sporangia

o resting spores

Note: Interbiotic rhizoidal system (g - m)

All pictures at X450


a
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The rhizoidal system consists of a main rhizoid which gets thicker by the progressive development of the sporangium, from which arises a single thin lateral rhizoid (Fig.56f &57c) The lateral rhizoid may branch further producing several smaller, thinner rhizoids which may not necessarily terminate in the host cells. Tapered tips of the lateral and sublateral rhizoids become attached to several algal cells, forming an interbiotic system (Fig.57g). Owing to the dense contents of the algal cells the internal rhizoidal system which probably exists was not observed.

Dehiscence was observed only once, when the zoospores emerged in groups (Fig.56k). Dehiscence was apical, zoospores were extruded through the apical pore which was formed by the detachment of the apex of the sporangium (Fig.56k). Thus the apex of the sporangium appeared to function as an operculum. It has not yet been established whether the species is operculate or inoperculate, however, the evidence so far suggests that it is operculate. Following the extrusion of the zoospores, the sporangium remains relatively intact (Fig.56m,n)however, a few may collapse to a certain degree (Fig.57 m-p).

The spherical zoospore (Fig.561) is 3 - 4μ in diameter and contains a single oil globule. The zoospore possess a single, posteriorly placed flagellum, 16μ in length.

The yellowish resting spore is spherical (Fig.566)11 - 13μ in diameter and surrounded by a thick-smooth wall. The cytoplasm is granular in appearance and does not appear to contain oil globules. No gametes were observed at any stage of the life cycle which may indicate that the resting spores were produced asexually, probably from sporangia.

Fig.57. Micrographs of developmental phases of a new chytrid species on Microcystis aeruginosa

a - h developing sporangia (→ indicates vacuole-like process)

i - l mature sporangia

m - p empty sporangia

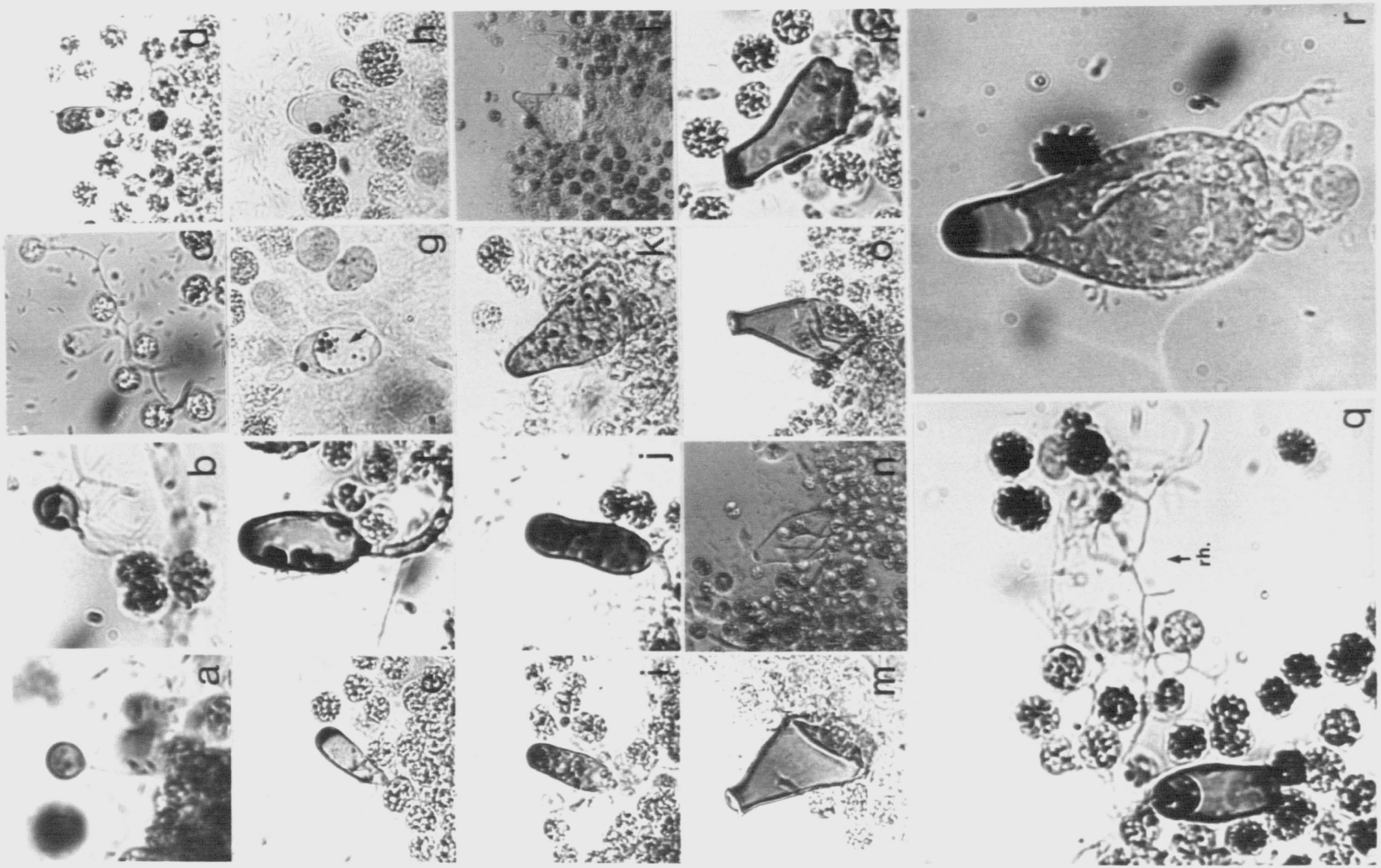
q interbiotic rhizoidal system

r immature sporangium (note the size)

c,d,e,i,l,n at X450

All others at X860

Note: The cells with greyish appearance are dead cells


a
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rh.

SKUJA (1948) has described a new chytrid, Chytridium microcystidis Skuja as a parasite of Microcystis aeruginosa. However, his form differs from the present chytrid in its rhizoidal system which consists of several rhizoidal axes. The sporangia of both chytrids appear to be similar in shape.

The present chytrid resembles species of Rhizophylctis and Polyphagus in its polyphagus habit. However, it differs from the former by virtue of its single-branched rhizoidal axis arising from the sporangium and from the latter in the enlargement of the zoospore directly into the sporangium. Its development appears to be similar to species of Dangeardia (Phlyctidiaceae) and Rhizidium windermerense Canter. Nevertheless, it differs from the former in respect of its interbiotic rhizoidal system which resembles the latter. The dehiscence and the resting spore of this chytrid seem to be similar to that of Rhizidium windermerense. In addition they are both polyphagus. Thus this chytrid overall appears to show the main characteristic features of the genus Rhizidium. The author therefore places this fungus into the same genus as a new species, Rhizidium microcystidis for the time being.

Parasitism and Epidemics

Development of this chytrid always commenced on healthy cells and usually on growing host populations. This suggests that the chytrid is a parasite.

The encysted zoospore sends a delicate germ tube into the mucilage surrounding the alga. A still unbranched or little

branched rhizoid grows inwardly and the growth continues until contact is made with a host cell. At this stage the infected cell is still healthy in appearance. A lateral branch often develops close to the sporangial rudiment and grows in more or less the opposite direction to that of the original axis. Therefore the thallus may have an interbiotic rhizoidal system, enabling the fungus to infect more than one cell. At this stage infected cells die; internally they appear relatively granular losing the characteristic features of a healthy cell (Plate 10).

The following table shows the distribution of developmental stages of chytrid during epidemics.

	1978					1979	1980			
Periods of epidemics	Sept 19	2	Oct 16	30	Nov 27	Oct 1	Sept 29	Oct 13	Nov 24	Dec 8
Germi.zoos		9	4			2	22	17	4	
Develop.sp	3	7	17	3	2	1	23	20		
Immat.sp.	11	2	34	4			83	12	4	2
Mature sp.	3	7	16				14	12		
Empty sp.		55	66	10			35	2		
Resting sp.							10			

Table 10 Developmental phases of the chytrid. Average on 20 colonies during epidemics.

It is apparent from the above table that numbers of developing sporangia were always more abundant than those of sporangia at the beginning of the epidemic. Developing sporangia were present until the last stage of infection showing a similarity to epidemics of Zygorhizidium affluens on Asterionella formosa. The highest numbers of sporangia coincided with the highest infections. During the epidemic in 1978, however, sporangia were far more abundant than during the epidemics of 1979 and 1980 although the latter was more severe (Fig.58). Resting spores were found only during the most severe epidemic (1980), thus supporting the dependency of resting spore formation on the degree of infection. This case was also very much similar to that of Z. affluens.

The chytrid showed a marked regularity in occurrence always coinciding with the presence of M. aeruginosa in Shearwater. Three epidemics were recorded in successive years (1978 - 80), all occurring regularly towards the end of September (Fig.58). Epidemics in 1978 and 1980 were quite severe, reaching 75% (1978) and even higher, 90%, in 1980. In fact the degree of infection (90%) in 1980 was the highest of all fungal infections, recorded on the phytoplankters of Shearwater. Both epidemics lasted over two months. The epidemic in 1979 was shorter and less severe (15%) in contrast to the other epidemics; the reason for this lower rate of infection is unknown.

The net sample often differed from the water sample. M. aeruginosa was quite abundant in the net samples from

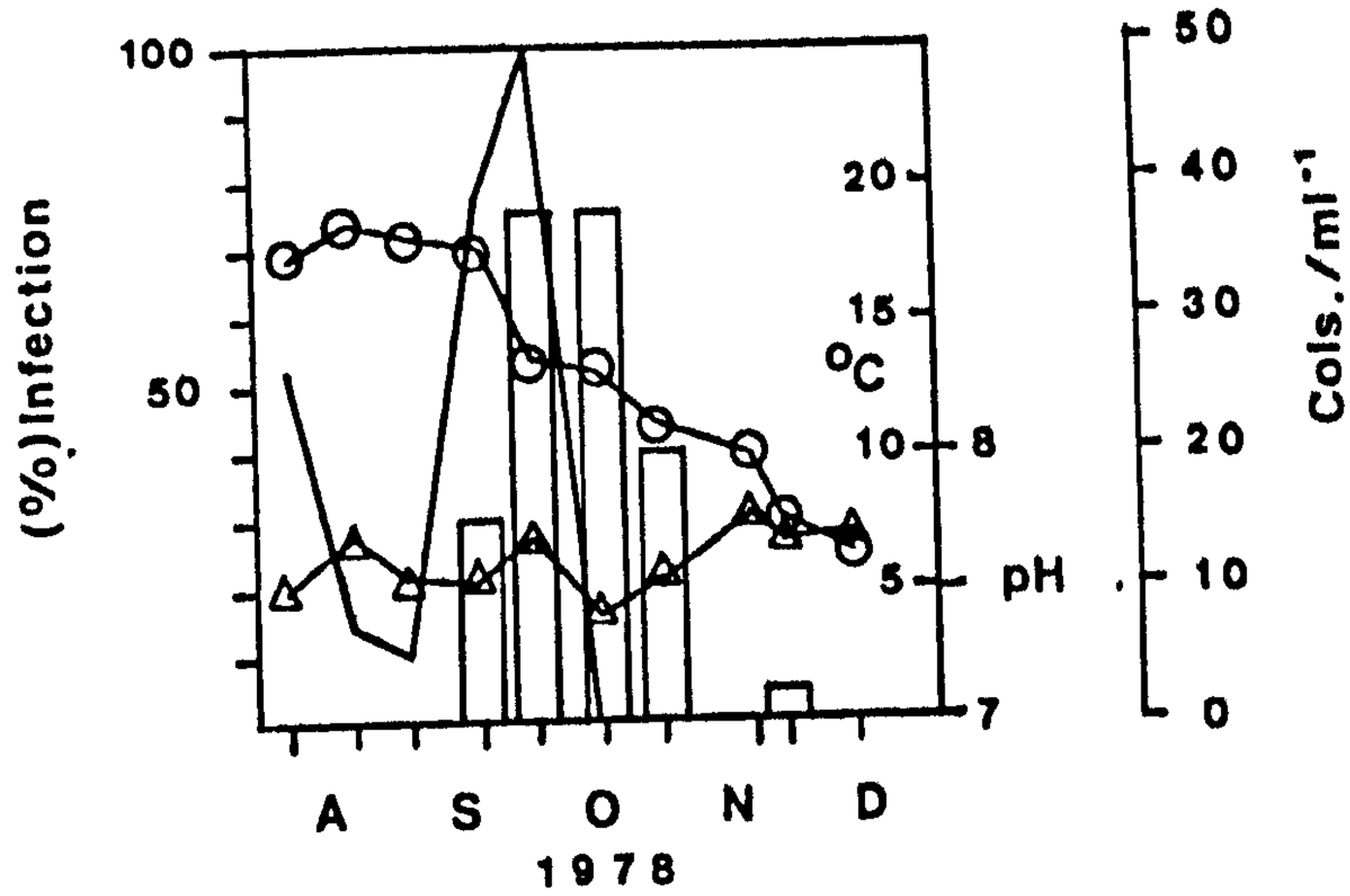
Fig.58. Epidemics of a new chytrid species parasitizing Microcystis aeruginosa in relation to physical factors.

□ % fungal infection

(———) number of cols. of M. aeruginosa/ml^{-1}

△——△ pH

○——○ temperature


100
50
(%)Infection
20
15
°C
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8
pH
5
7
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10
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Cols./ml⁻¹
A
S
O
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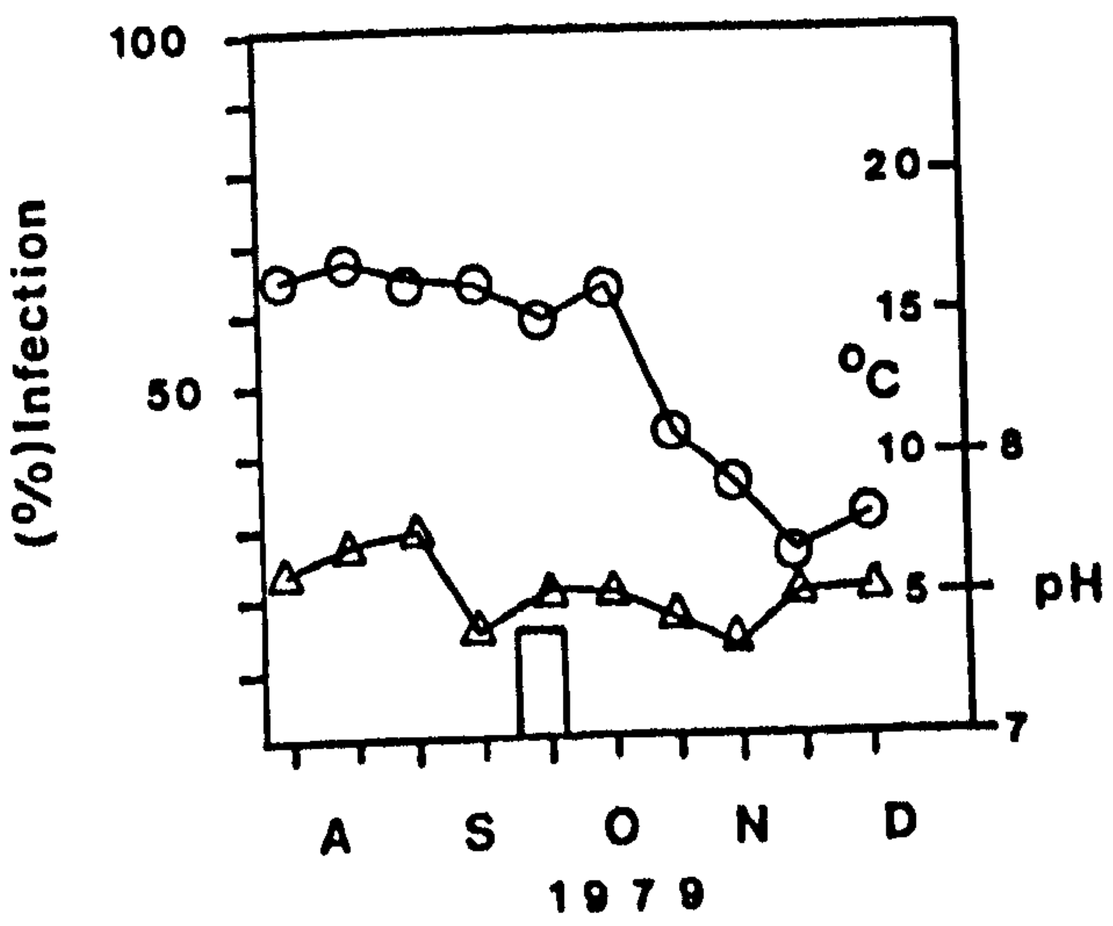
1978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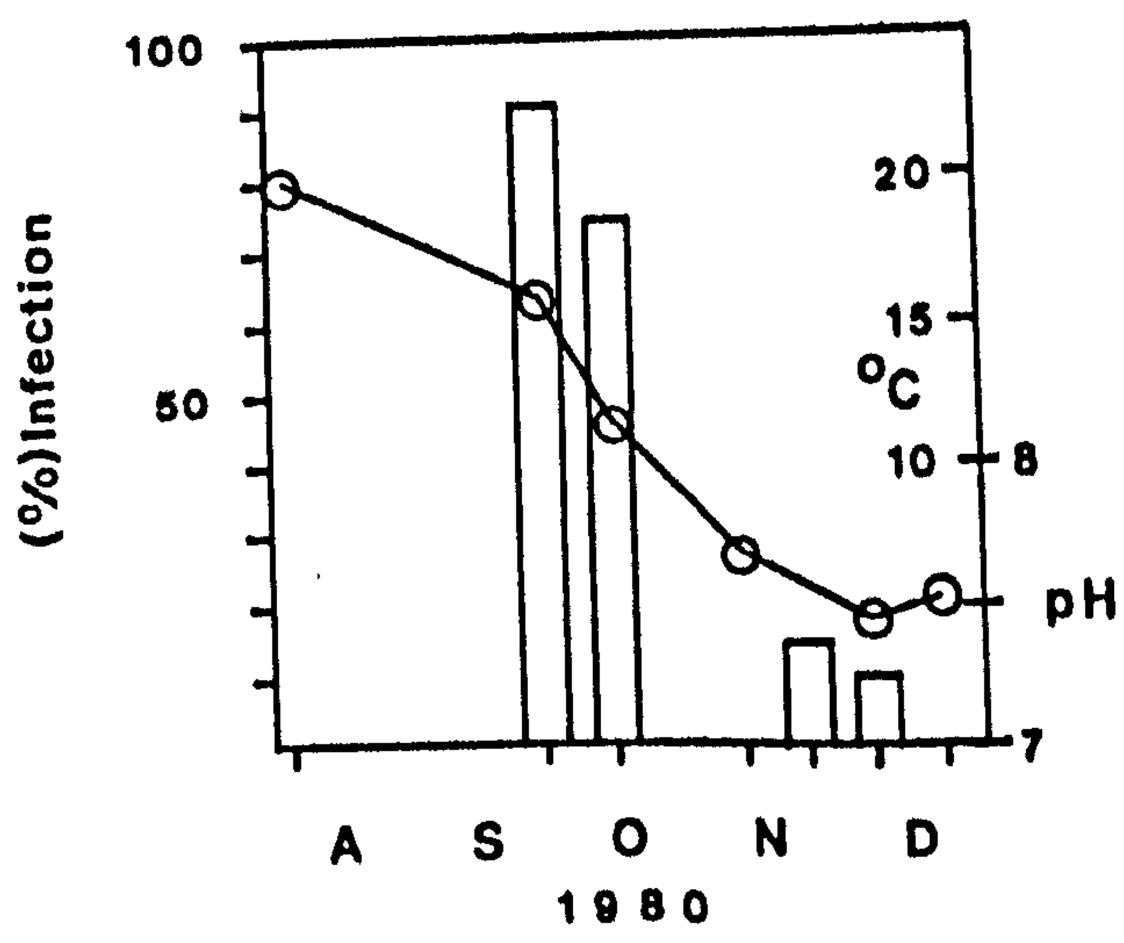
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(%)Infection
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pH
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A
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O
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D
1980

which the infection was recorded whilst the water sample often contained very few colonies. In addition other similar colonial types of the blue-green algae such as Coelosphaerium naegelianum and Gomphosphaeria naegeliana usually occurred in the same period. This made it quite difficult to count the individual colonies in the water samples. Numbers of colonies of these blue-green algae, therefore, are shown as totals in Fig.19.

Despite the difficulties during the epidemic in 1978, I was able to count colonies of M. aeruginosa separately; these are shown in Fig.58.When the epidemic started the number of colonies was increasing and the rise continued until the maximum infection (75%) was reached. Shortly after this stage the alga disappeared from the water sample. Nitrogen but not phosphorus was shown as a limiting factor for the growth of M. aeruginosa in the lakes of Southern Wisconsin by GERLOFF & SKOOG (1954, 1957). During this epidemic the concentrations of both nitrate and silicate were either increasing or actually high in Shearwater (Fig.3) At this stage water level was low thus limiting the effect of floods for the disappearance of M. aeruginosa. VANCE (1965) showed by quantitative culture techniques that M. aeruginosa is vulnerable to a substance liberated by Euglena sp. which was never found in Shearwater. Zooplankters were also absent during this period in the present study (Fig.28). Temperature was quite high and thus in favour of M. aeruginosa (Fig.58) However, there was a decrease in pH which then increased afterwards though the alga did not appear in Shearwater (Fig.58)

Limitation of growth of the blue-green algae around pH 6.5 and good growth over ph 7 is well known. In the present study pH value never dropped under 7.3 at any stage during this investigation (Fig. 1). The end of the thermal stratification gives rise to turbulence and this might possibly be another reason for the disappearance of the alga during this epidemic. Eventually parasitism appeared to be one of the factors responsible for the disappearance of M. aeruginosa in 1978.

Factors affecting the occurrence of the chytrid

Temperature and pH appeared to be in a way governing the occurrence of this chytrid in Shearwater (Fig.58). Two main epidemics started when temperature was over 15^{o}C and the end of the epidemics coincided with much lower temperatures (around 5^{o}C). In 1979, during the very short appearance of the chytrid, although the temperature pattern was almost the same, development of the chytrid however did not continue. pH also underwent a similar pattern of change (7.4 - 7.7) during the first two epidemics (data is not available for the last epidemic).

The onset of epidemics coincided with rising water level indicating the increase in the nutrional composition of the lake. The concentrations of nitrate, phosphate and silica were either increasing or high during epidemics (Fig. 3). Therefore these nutrients may appear to be important factors governing the onset of epidemics. In conclusion the present data would suggest that a group of factors appears to govern

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the occurrence of this chytrid.

This chytrid was not observed on any other planktonic algae in Shearwater. C. naegelianum and Gomphosphaeria naegelina were particularly present in the same period but they remained fungus-free. This is a good example of host specifity for this chytrid. In addition the chytrid appeared only when the host M. aeruginosa was present, showing the importance of suitable host availability for the growth of the fungus.

Summary and conclusions

A new chytrid, Rhizidium microcystidis has been described as a fungal parasite of the blue-green alga, Microcystis aeruginosa Kuetz.; emend. Elenkin. in this section.

The chytrid occurred quite regularly and its occurrence showed a clear relationship with physico-chemical factors, studied during this study.

The chytrid showed a great host specifity, growing only on M. aeruginosa although other colonial blue-green algae were present in the periods of parasitism.

Infection commenced with a high number of developing sporangia and ceased with high numbers of sporangia. The highest number of sporangia coincided with the most severe epidemic.

Resting spores were found only during the most severe epidemic, indicating the dependency of resting spore formation on the degree of infection.

Maximum infection of 90% was also the overall highest of all the fungal infections recorded in this study.

Parasitism appeared to be one of the factors responsible for the disappearance of M. aeruginosa in Shearwater.

Parasitism caused an increase in the number of dead cells of the alga.

CHAPTER 4.

THE OCCURRENCE OF COLOURLESS FLAGELLATES EPIPHYTIC ON PLANKTONIC ALGAE IN SHEARWATER.

The occurrence of colourless flagellates, Bicosoeca lacustris James-Clark (Bicosoecales) and choanoflagellates, Codosiga botrytis (Ehr.) Saville-Kent and species of the genus Salpingoeca (James-Clark) Saville-Kent (Craspedomonadales) on certain planktonic algae was quite striking in Shearwater, displaying an interesting distribution on different planktonic algae.

Members of the genus Bicosoeca and choanoflagellates are common in many freshwater bodies and also occur in brackish and marine environments. The species of the genus Bicosoeca James-Clark live in ponds and lakes mostly attached to the submersed plants and animals and often to the free-floating protists while choanoflagellates are common members of the nannoplankton and generally regarded as neuston organisms.

The studies on these flagellates have consisted mainly of taxonomic descriptions or more recently ultrastructural investigations and their ecology has been somewhat neglected. NORRIS (1965) found choanoflagellates attached to the air-water interface in an inverted position while BOUCAUD-CAMOU (1966) obtained the specimens from the surface of algae, hydroids and bryozoans but noted that they occurred in very small numbers. The choanoflagellates observed by LACKEY (1967) were mainly planktonic. HILLIARD (1971) reported the occurrence of many species of the genus Bicosoeca on planktonic algae (blue-green, diatoms and chrysophyceae) showing a wide distribution of

the flagellates on various algae. His study also concentrated on the taxonomic description of the flagellates. HIBBERD (1975) studied the ultrastructure of Codosiga botrytis, which was found attached to the diatom Asterionella formosa, neglecting the occurrence of the flagellate on the diatom.

Investigations on the occurrence of Bicosoeca lacustris and choanoflagellates, Codosiga botrytis and Salpingoeca spp. in Shearwater has clearly demonstrated that these flagellates are common in the plankton and may have a distinct distribution on planktonic algae. Their ecology, therefore, has been studied quantitatively.

Method

Separate counting of individual flagellates on or around the planktonic algae was made on fresh net samples and their numbers and percentage (%) attachment were determined by counting 60 - 100 colonies of blue-green algae and/or filaments of diatoms.

The flagellates were drawn by using camera lucida at X 450 magnification. The light micrographs were obtained with an Olympus BH1 photomicroscope using PAN 35mm films. The differential interference contrast optics were used in most cases for the morphological details of the flagellates and the flagellates were stained by neutral red in some instances. The flagellates were fixed in 5% glutaraldehyde for T.E.M. studies. The samples were then washed in distilled water for 5 - 6 times and a drop was then placed on a copper grid with a plastic film of Formvar with a micropipette. After allowing the drop of

sample to dry, it was shadowcoated with platinum carbon evaporation. The grid was then examined with A.E.I. EMGG Transmission electron microscope.

The genus Bicosoeca James-Clark

The genus Bicosoeca James-Clark comprises a rather obscure group of species of freshwater and marine colourless flagellates, and has been placed both in the algae and in the protozoa, hence this has confused an already difficult systematic situation.

The genus was erected by JAMES-CLARK (1868) to include two species. JAMES-CLARK derived the name Bicosoeca from the Greek βΐκos, a vase and olkéw, to inhabit. The generic name was then changed to Bikoeca by STEIN (1878). The philogically preferable compound would be Bicoeca and most subsequent authors have followed STEIN (1878) and "corrected" the original spelling. However, according to Article 73 of the International Code of Botanical Nomenclature (STAFLEU, 1972), the original spelling of the name cannot be considered incorrect and it must be used in its original form. If the organism is classified as an animal the same conclusion is confirmed by application of Article 32 of the International Code of Zoological Nomenclature (STOLL, 1964). Therefore the original generic name Bicosoeca will be used throughout this paper.

Species of Bicosoeca are biflagellate; solitary or colonial body transparent; the protoplasm is small, surrounded by a characteristic vase-like lorica; the shorter posterior flagellum acts as an attachment organelle; the longer flagellum is a swimming organelle; one nucleus and one or two contractile vacuoles; holozoice; reproduction is by transverse fission; sessile or free-swimming; mostly freshwater,also brackish or marine.

The taxonomy of the genus has been based mainly on the morphology and structure of the lorica and its taxonomy has always been disputable since its establishment over a hundred years ago by JAMES-CLARK (1868).

Several authors, from JAMES-CLARK (1868-69) onwards place the group near the Craspedophyceae, or even in Craspedomonadales (SKUJA, 1956), since the apical 'lip' has been considered equivalent to the craspedophycean collar. However, it has been shown (PETERSON et al. 1954) that the craspedophycean collar consists of individual tentacles, hence there is no reason to suggest such a close relationship. There are also no close similarities in lorica structure since the Bicosoeca lorica has a complicated fibrillar system whereas the craspedophycean lorica appears amorphous (KRISTIANSEN, 1972).

KLUG (1936) tentatively assigned Bicosoeca to the Chrysophyceae on account of the endogenous cysts which he studied in Bicosoeca lacustris James-Clark. FOTT (1946) has observed a similar cyst in B. mitra Fott and WILLEN (1963) in B. cylindrica (Lackey) Bourrelly which is spherical with a thick verrucose wall but no porus, lying free within the lorica. However, KRISTIANSEN (1972) has pointed out that more observations are needed to determine whether these are endogenous cysts formed as in Chrysophyceae or whether a porus is present. Since many genera have similar statospores (cysts) it may not be valid to use them as diagnostic features (TRAINOR, 1978). If of typical Chrysophycean form then they can be used diagnostically to distinguish certain genera of the class. When each cyst form is connected to a species then they may be able to be used as diagnostic of species and even of genera if each genus has some particular cyst form.

The genus Bicosoeca has been placed among Protomonadales as "colourless flagellates of uncertain position" (PICKEN, 1941 and FOTT, 1959). HUBER-PESTALOZZI (1941) placed the genus in the family Bicosoecaceae (Protomastiginae), which also included one other genus, Histiona.

PASCHER (1943) removed colonial species from Bicosoeca and placed them in a separate genus Stephanocodon whereas BOURRELLY (1951) divided the genus Bicoseoca into four sections: Eubicosoeca (epiphytic), Codomonas (planktonic, solitary), Stephanocodon (planktonic, colonial), and Poteriodendron (sessile, colonial). Although GRASSE & DÉFLANDRE (1952) believed that Bicosoeca is reminiscent of the Chrysophyceae on account of general structure, they also suggested a relation to Bodo because of a similar function of the posterior flagellum as an organelle of adherence and errected four completely different genera based almost on the sections of BOURRELLY (1951); Bicosoeca James-Clark, Codomonas Lackey, Poteriodendron Stein and Stephanocodon Pascher. FOTT (1959, 1960) is in accord with GRASSE & DÉFLANDRE. More recently KUDO (1966) has extended the family Bicosoecaceae to include some genera of questionable status: Bicosoeca James-Clark, Salpingoeca James-Clark, Codonoeca James-Clark, Diplosigopsis Francé, Histiona Voigt, Proteriodendron Stein and Lagenoeca Kent. Finally, BOURRELLY (1968) recombined them into one genus. However, KRISTIANSEN (1972) has suggested that the existence of solitary or colonial species should not in itself justify the splitting up into separate genera although he allowed that the fundamental differences in lorica structure might necessitate a division

of the genus. Although HILLIARD (1971) found that the loricae of many species show distinct striations, recent data from electron microscope studies have revealed an unsuspected range of variation among species which have been thought to have undifferentiated loricae (HIBBERD, 1978).

GRASSE & DÉFLANDRE (1952) were the first biologists after KLUG (1936) to consider the genus Bicosoeca as reminisent of the Chrysophyceae but in conclusion they pointed out that no single criterion is sufficient in itself to relate the genus conclusively with the Chrysophyceae. They moreover felt that an investigation of the flagellum could decide this question. BOURRELLY (1968) has placed the Bicosoecales as an appendix to the Chrysophyceae. More recently, HILLIARD (1971) examined many species of the genus in detail using the staining technique of Vilh.Jensen and found two criteria which may firmly relate Bicosoeca with Chrysophyceae: the presence of mastigonemes on the anterior flagellum and the nature of the lorica.

The flagellar structure is an argument for placing the Bicosoecales near the Chrysophyceae and the features of lorica structure are comparable to those known from Chrysophyceae (BELCHER, 1968; KRISTIANSEN, 1969). MIGNOT (1974) examined the ultrastructure of Bicosoeca lacustris and B. kepneri and found that the ultrastructural organisation of Bicosoeca is different from the basic structure of Bodo and Choanoflagellates, but that it shows many affinities with the Chrysophyceae (anterior flagellum of Bicosoeca bears mantigonemes, which are tubular and of the same type as those of the Chrysophyceae).

MOESTRUP & THOMSEN (1976) agree with MIGNOT (1974) that Bicosoeca is related to the Chrysophyceae but also stress that there are features in Bicosoeca without any known equivalent in the Chrysophyceae. They also add that without a more detailed knowledge of the flagellar apparatus generally in the heterokont flagellates, it is difficult to make definite statements. KRISTIANSEN (1972) speculates that only the discovery of a pigmented relative would finally settle the problem, but until that time, they should be regarded as a separate class in view of their very specialised flagellar apparatus. In addition, HIBBERD (1977a) also suggested that the genus Bicosoeca should be considered separately until a new order paralleling that for the choanoflagellates is created.

It would be quite desirable to add some more points to the taxonomic position of the genus Bicoseoca through the present study, however this must be considered as a future project since occurrence of a single Bicosoeca species in Shearwater somewhat limits this.

Bicosoeca lacustris James-Clark

The overall shape of a mature complete cell is more or less pyriform (Fig.59a), slightly rounded posteriorly, broadest in the middle and truncate anteriorly measuring 18 - 19µm from the tapered anterior to the end of the posterior flagellum. The lorica is elongate-ovate, widest posteriorly, 10 - 14µm long, 5 - 5.5µm broad, a little less than twice as long as the body. The cell body is small, more or less pyriform in shape, 8µm long, 5µm broad (broadest point). The cell body lies in a characteristic vase-shaped lorica and oontains a generally centrally placed nucleus and 1 - 2 posterior contractile vacuole. At some stages the cell body was observed to fill the whole lorica (Fig.59b) or come out of the lorica and sit on it (Fig.59c). These stages are probably associated with the reproduction of the species. The tapered anterior of the cell body has a laterally projecting trophic flagellum, 23 to 26µm long at fully stretched position. Coiling, stretching and contraction stages of the anterior flagellum are illustrated in figure 59a-c . Transmission electron micrographs of B. lacustris in this study revealed that the longer anterior flagellum bears mastigomes on both sides (Fig 60a-c) thus supporting the finding of HILLIARD (1971). The presence of mastigomes on the anterior flagellum may relate B. lacustris to the Chrysophyceae whose members also have mastigomes on the flagellum. According to JAMES-CLARK (1868) the anus is also placed anteriorly on the lip-like process.

The biflagellate protoplast is attached to the base of the lorica by means of the tip of the shorter posterior

Fig.59. Bicosoeca lacustris James-Clark

(a - c) different appearances of cell body, lorica and flagella of B. lacustris

(d) Attachments of B. lacustris to planktonic algae.

All pictures at X450

a.fl = anterior flagellum

nc = nucleus

vc = contractile vacuole

lr = lorica

p.fl = posterior flagellum

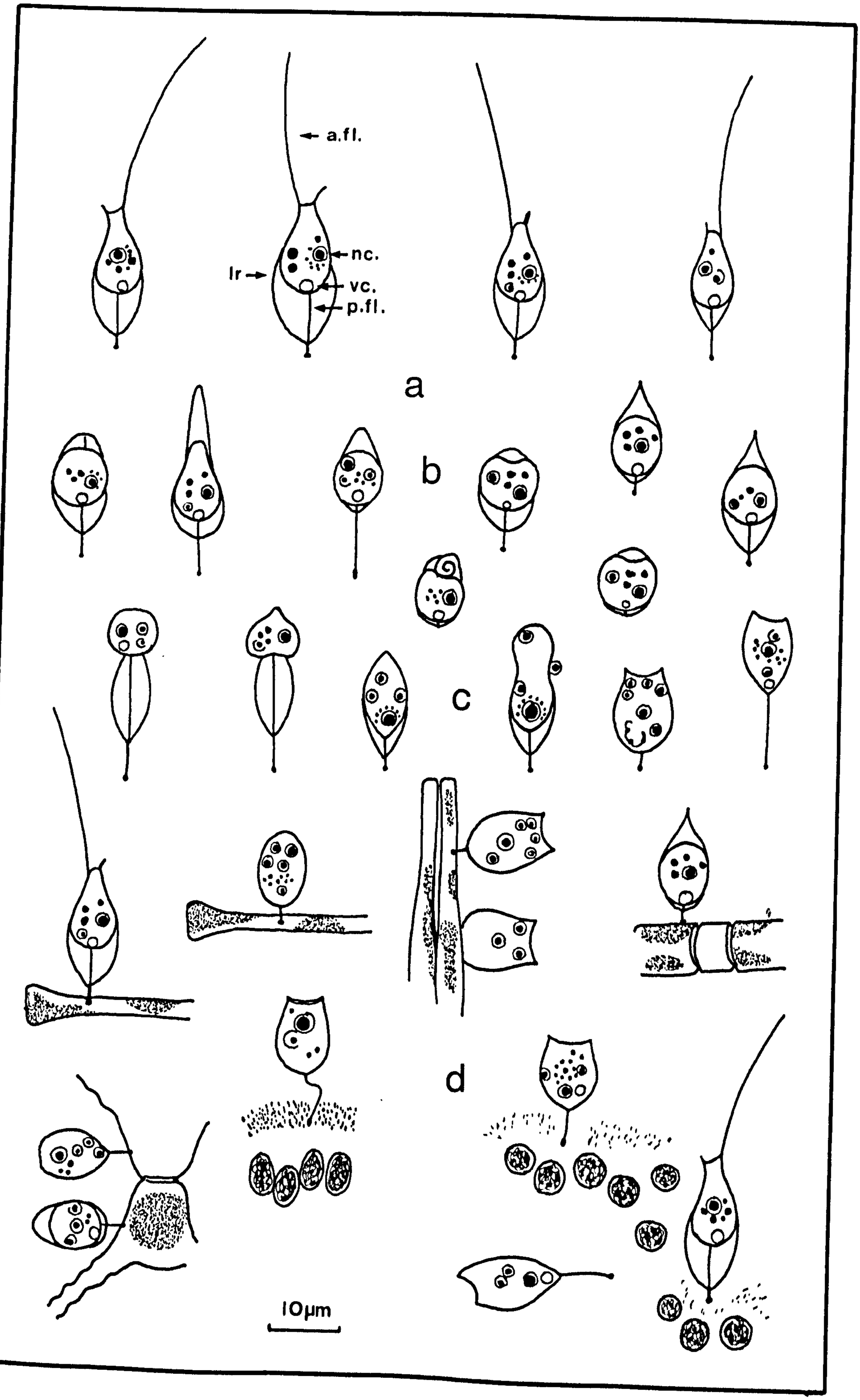

a.fl.
nc.
lr
vc.
p.fl.
a
b
c
d
10µm

Fig.60. T.E.Micrographs of Bicosoeca lacustris

a B. lacustris with anterior flagellum (note the flipper hairs on the flagellum) X2500

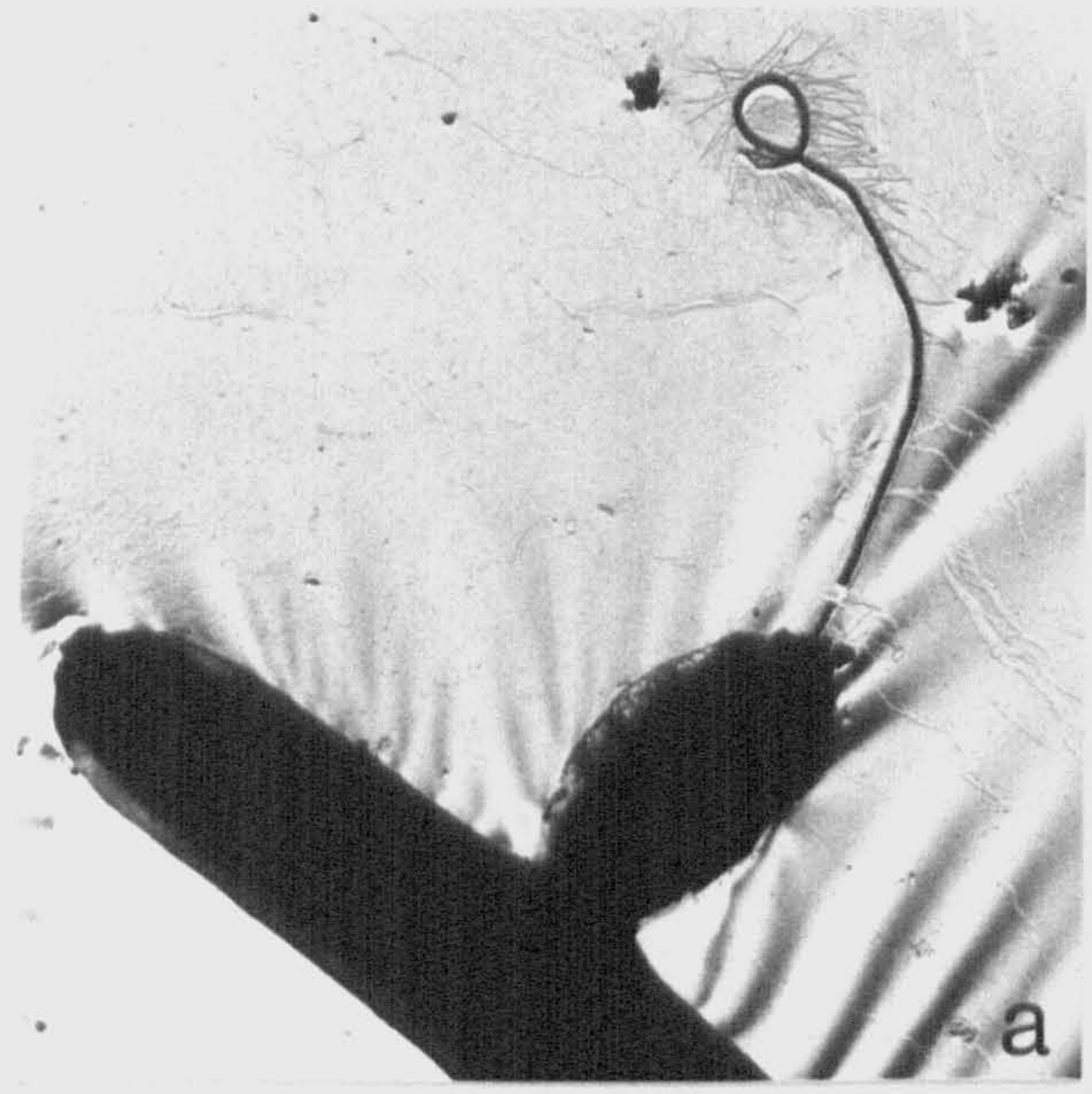
b Details of flipper hairs (mastigomes) X13000

c Whole lorica with anterior flagellum (note originating point of the flagellum and slipper hairs) X4000

d Posterior flagellum (note the attachment point of posterior flagellum X4000

e B. lacustris on F. crotonensis X2500

f B. lacustris on A. formosa X2500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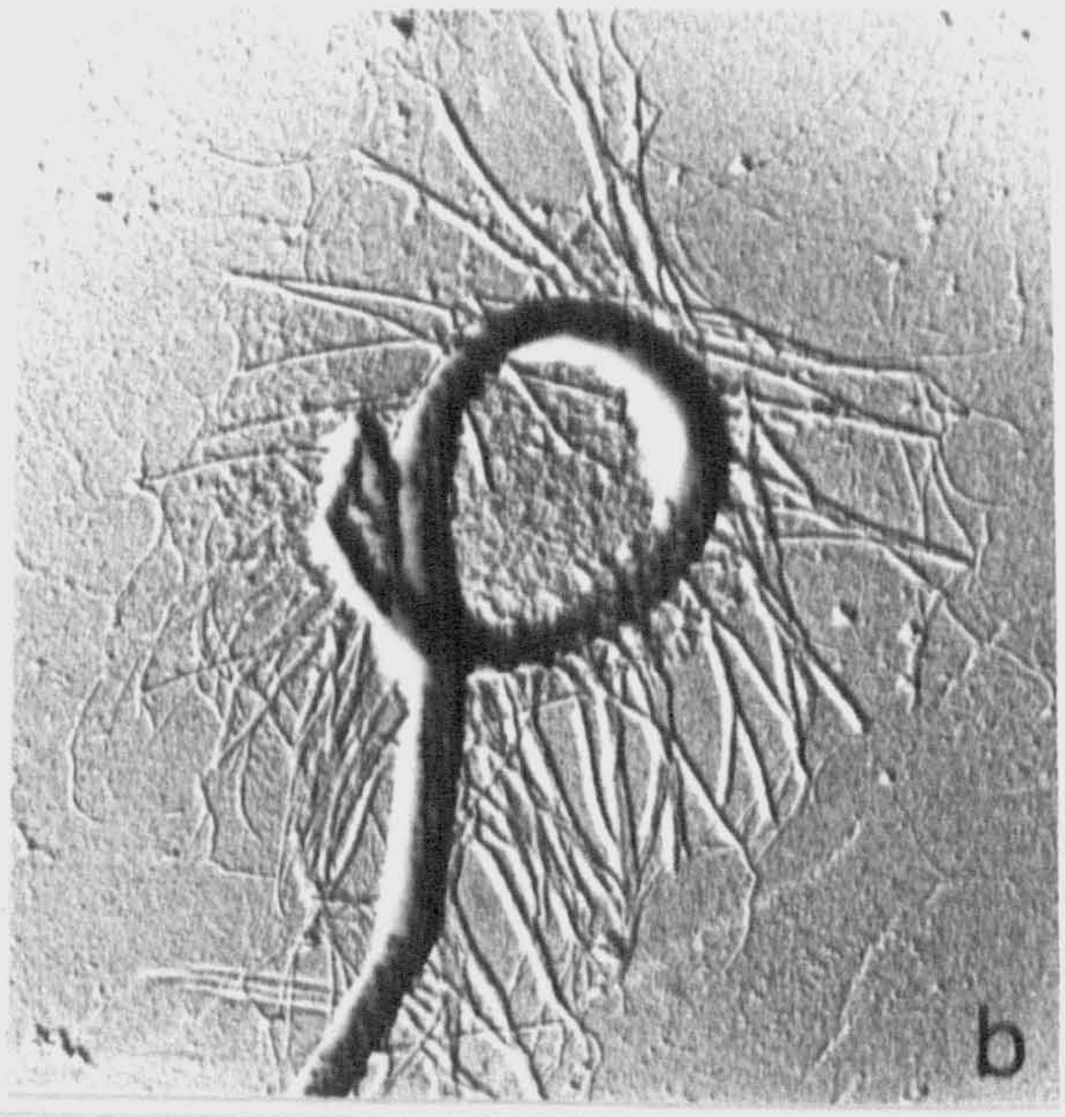
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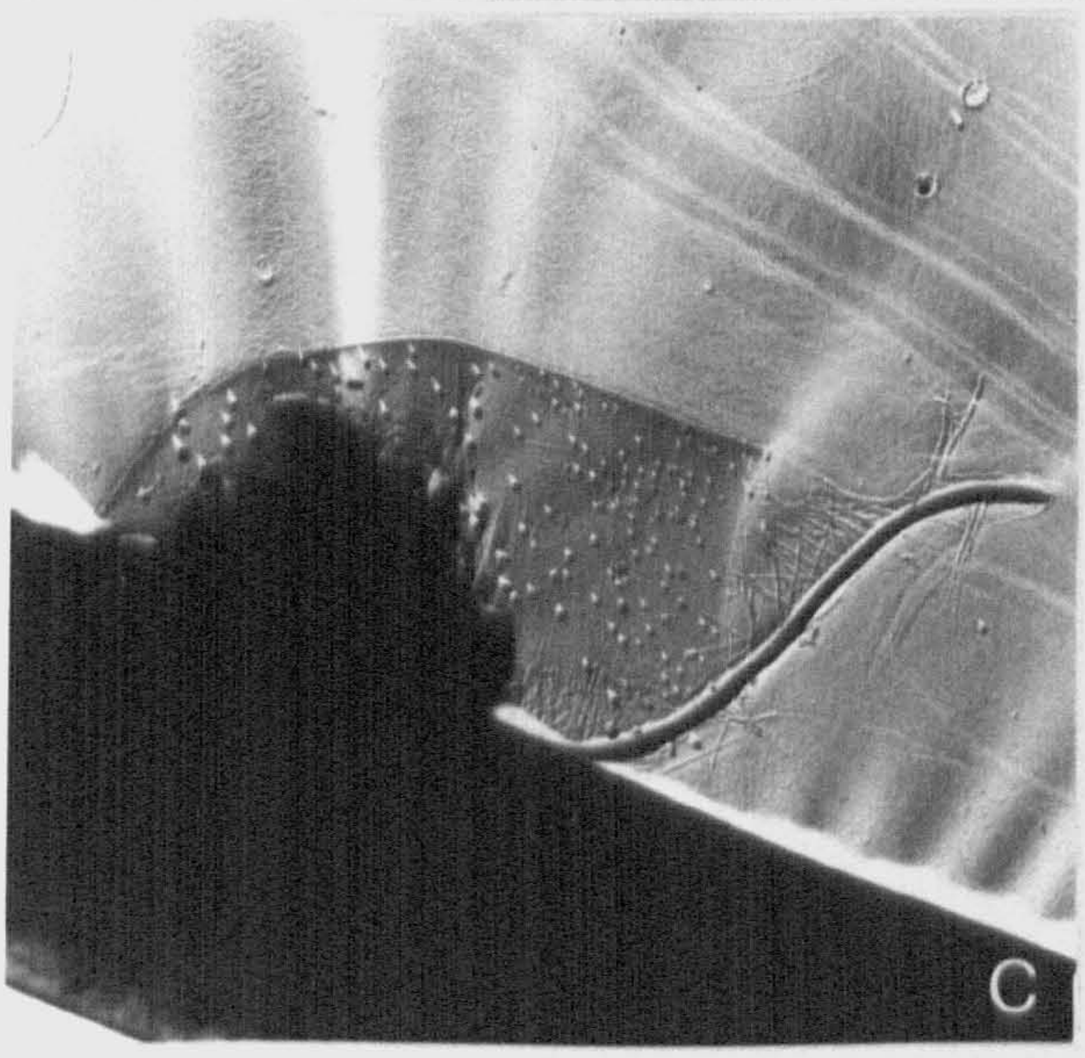
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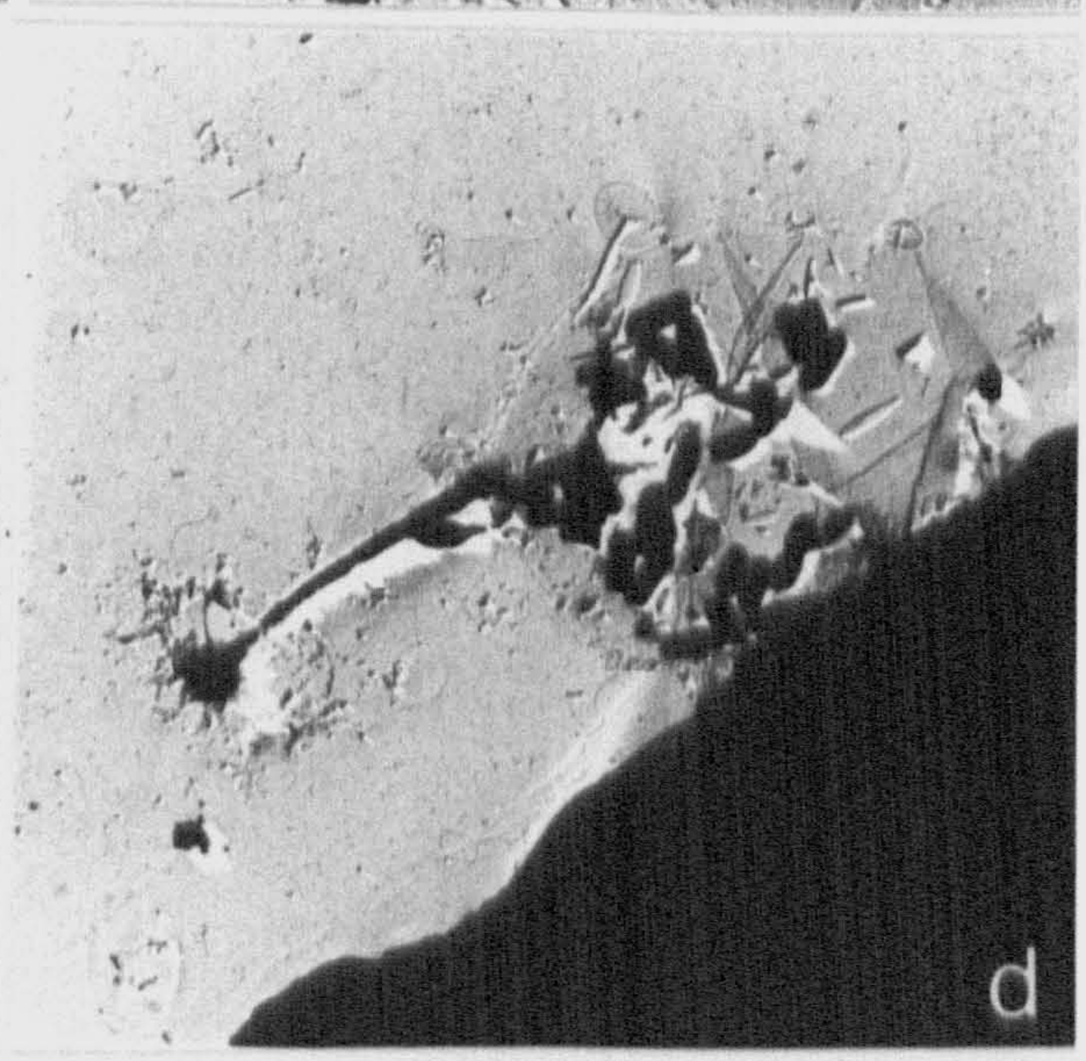
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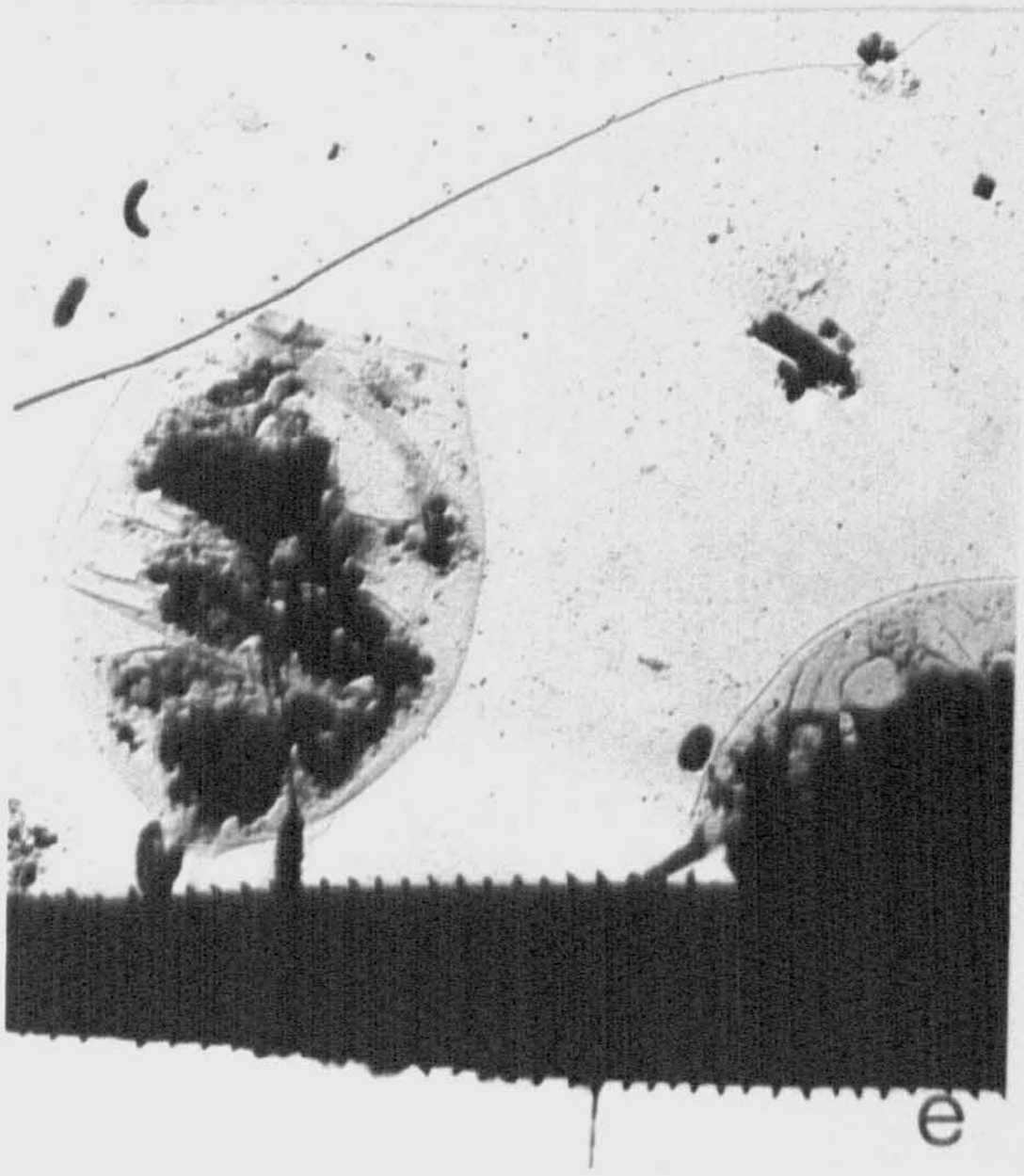
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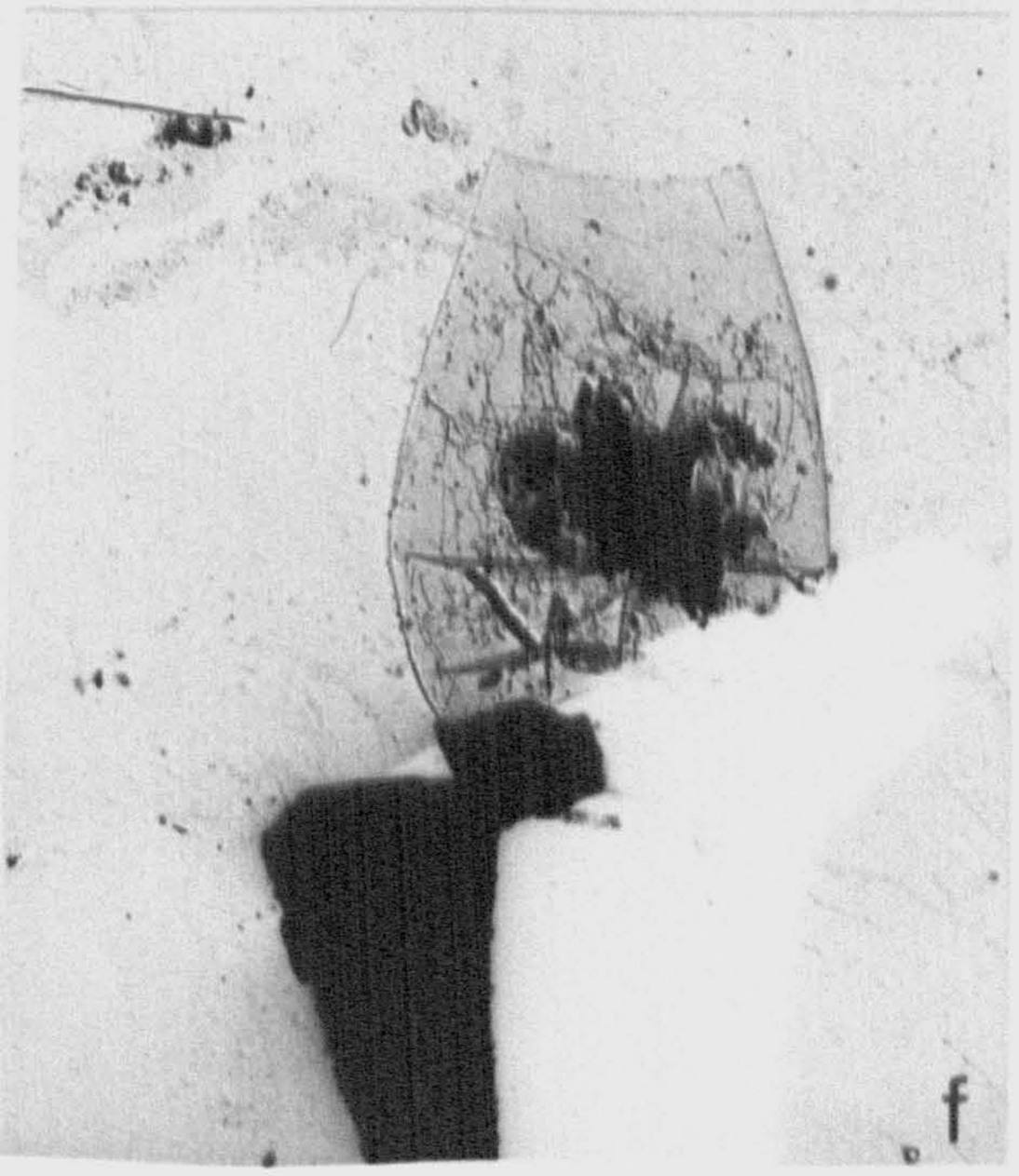

f

flagellum, 10 - 10.5µm long. Posterior flagellum is smooth without appendages (Fig.60d) and acts as an organelle of adherence. The posterior flagellum appeared to bend (Fig.59d). but was never observed to be contracted. A slight distal swelling appears to function as an hapteron (Fig. 60d). The real length of the posterior flagellum was observed when the cell body was out of lorica (Fig.59d); it is twice as long as when the cell is in the lorica. The length of the posterior flagellum at this stage measures 18µm indicating that the posterior flagellum originates from near the anterior end of the cell body. At the stage when the cell body fills the lorica the anterior flagellum is usually withdrawn over the lorica or actually separated and the posterior flagellum appears to be very short (Fig.59b,a). The similar stages of B. lacustris were also observed by JAMES-CLARK (1868) and KLUG (1936) during reproduction of the species. The method of reproduction in B. lacustris was not observed during this study but was reported to be a multiplication by transverse fission (for details, see KENT 1880-81 p.275-276 and KLUG 1936).

Craspedomonadales (Choanoflagellates)

The choanoflagellates are a distinct group of colourless flagellates characterised by the presence of a truncate collar extending out at the anterior end of the cell forming a ring around an apical flagellum. BOUCAUD-CAMOU (1966) divided choanoflagellates into three families; Codonosigidae, Salpingoecidae and Acanthoecidae. Members of only the first two families occurred in Shearwater. The family Codonosigidae includes species with or without lorica; stalked or stalkless occurring as solitary as well as colonial. The species of Salpingoecidae occur solitary and cells lie in a lorica made of chitin or cellulose. Family Salpingoecidae is more homogeneous than the Codonosigidae and members of both families could occur either attached to subrates or free in marine, brackish or freshwater environments.

The taxonomic position of the choanoflagellates has been subject of much interest and debate and these organisms still have a place in classifications of both the plant and animal kingdoms. In the system of BOURRELLY (1968) they form a subclass in the Chrysophyceae (golden-brown flagellate algae) and the Class Craspedophyceae in the Division Chromophyta (CHRISTENSEN, 1962, 1966) while they are included in the order Choanoflagellida in the Class Zoomastigophorea, Protozoa (HONIGBERG et al. 1964). More recently, PARKE & DIXON (1976) have deleted the Craspedomonadales from their third check-list of British algae.

Codosiga Botrytis (Ehr.) James-Clark

C. botrytis was the only representative of the genus Codosiga occurring in Shearwater.

Codonosiga (Codosiga) botrytis was erected by EHRENBERG under the genus Epistylis and referred to a new genus Codosiga by JAMES-CLARK (1866).

The cell body is normally more or less oval in outline and the majority of cells are 8 - 12µm long and 6 - 8µm wide (Fig.61v-x). The body is enclosed in a delicate, transparent close-fitting case which is continued below into a rigid stalk (Fig. 61v) by which the organism is attached to a substratum (algae)upon which it lives. The stalk varies a good deal in length probably according to the age of the individuals, measuring between 18 to 28µm. The posterior ends of cells occurring in pairs or fours, fuse into a single stalk at a distance of approximately 3 - 4µm from the cell body (Fig.61w-x).

A single flagellum arises from the middle of the anterior end of the cells and its average length is 26 - 30µm (Fig.61v) The flagellum is encased by a funnel-shaped collar (Fig. which is made up of 25 - 30 equally spaced cylindrical tentacles (Fig.62a,b) The length of collar is approximately 9 - 10µm. The flagellum gets thinner towards the distal end (Fig. 62a). The longer thick part of the flagellum seems to consist of lateral fibris (Fig. 62c) apparently indicating the fibrillar nature on the flagellum which is also observed by PETERSEN & HANSEN (1954) and HIBBERD (1975). The presence of a sheath around the flagellum has first been observed by

Fig.61. a - u different appearances of Salpingoeca spp.

v - x Codosiga botrytis

all pictures at X450

fl = flagellum

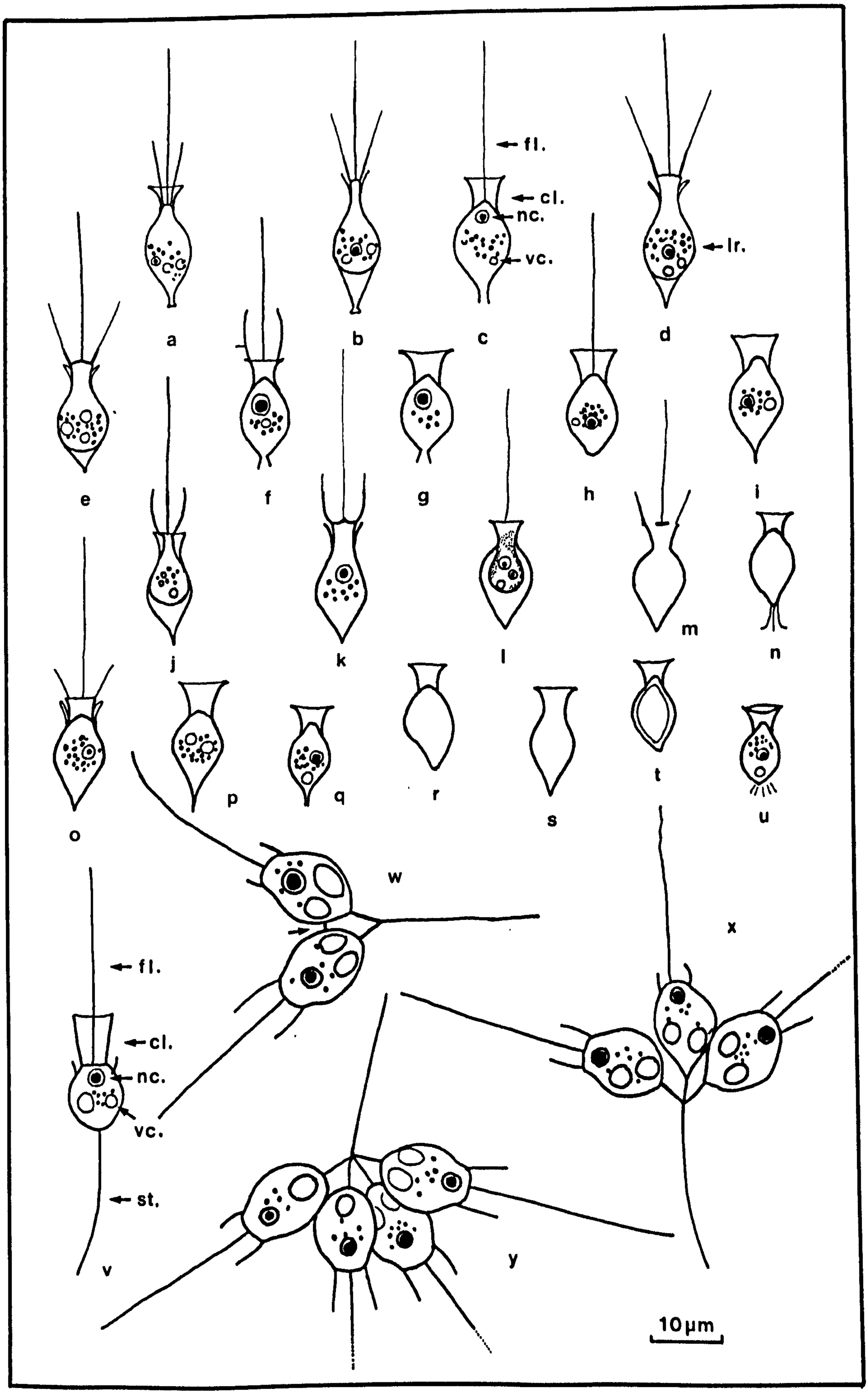
cl = collar

nc = nucleus

vc = vacuole

st = stalk

(→) in picture (w) indicates the cytoplasmatic bridge between two cells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10µm

PETERSEN & HANSEN (1954) and it was confirmed by the present study (Fig. 62c).

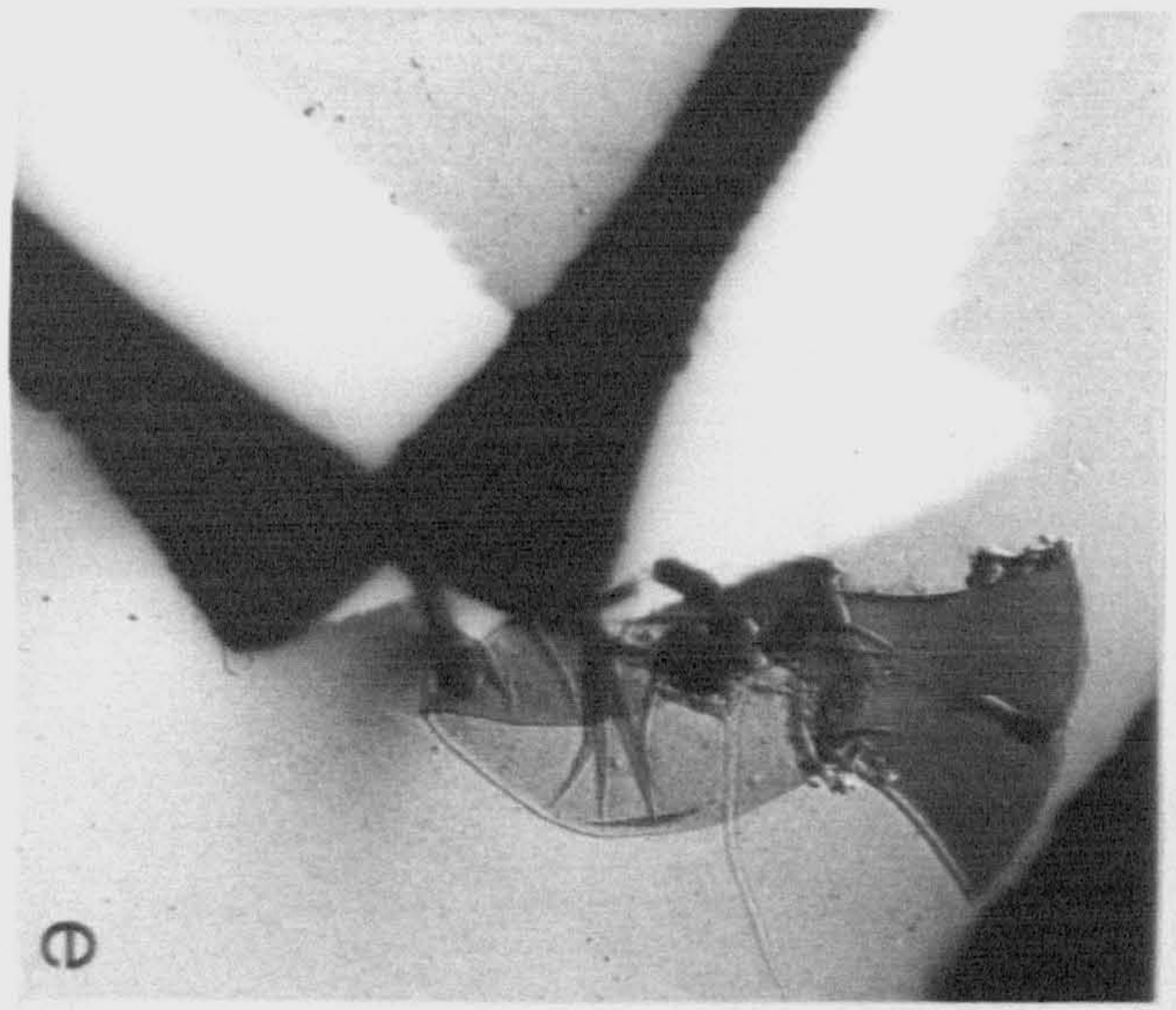
The internal structure of the cells appears to be relatively simple. The most conspicuous organelle is the spherical nucleus which measures 2 - 3µm in diameter, lying centrally near the anterior end of the cell (Fig.61v-x). The protoplasm contains 1 to 3 large vacuoles which are situated near the posterior end of the body.

The cells occurring in pairs are not held together only by the stalk but by a lateral linkage of the cells bodies towards their posterior ends by a cytoplasmic bridge which is hardly seen in the light microscope (Fig.61w), thus supporting the finding of HIBBERD (1975).

The method of reproduction was not observed during this study, however according to earlier workers the organism multiplies by binary fusion, the nucleus, body and collar all being split into two after retraction of the flagellum.

Fig.62. T.E.micrographs of Codosiga botrytis (a - d) and Salpingoeca sp. (e - f).

a	collar and anterior flagellum	X4000
b	collar at higher magnification showing the tectacles	X13000
c	middle part of the anterior flagellum (→)indicates the presence of hairs on the flagellum.(Also note the sheath of flagellum	X20000
d	A pair of C. botrytis	X4000
e	Salpingoeca sp. on Asterionella formosa (note: few striations on the lorica)	X2500
f	Amorphous collar of Salpingoeca	20000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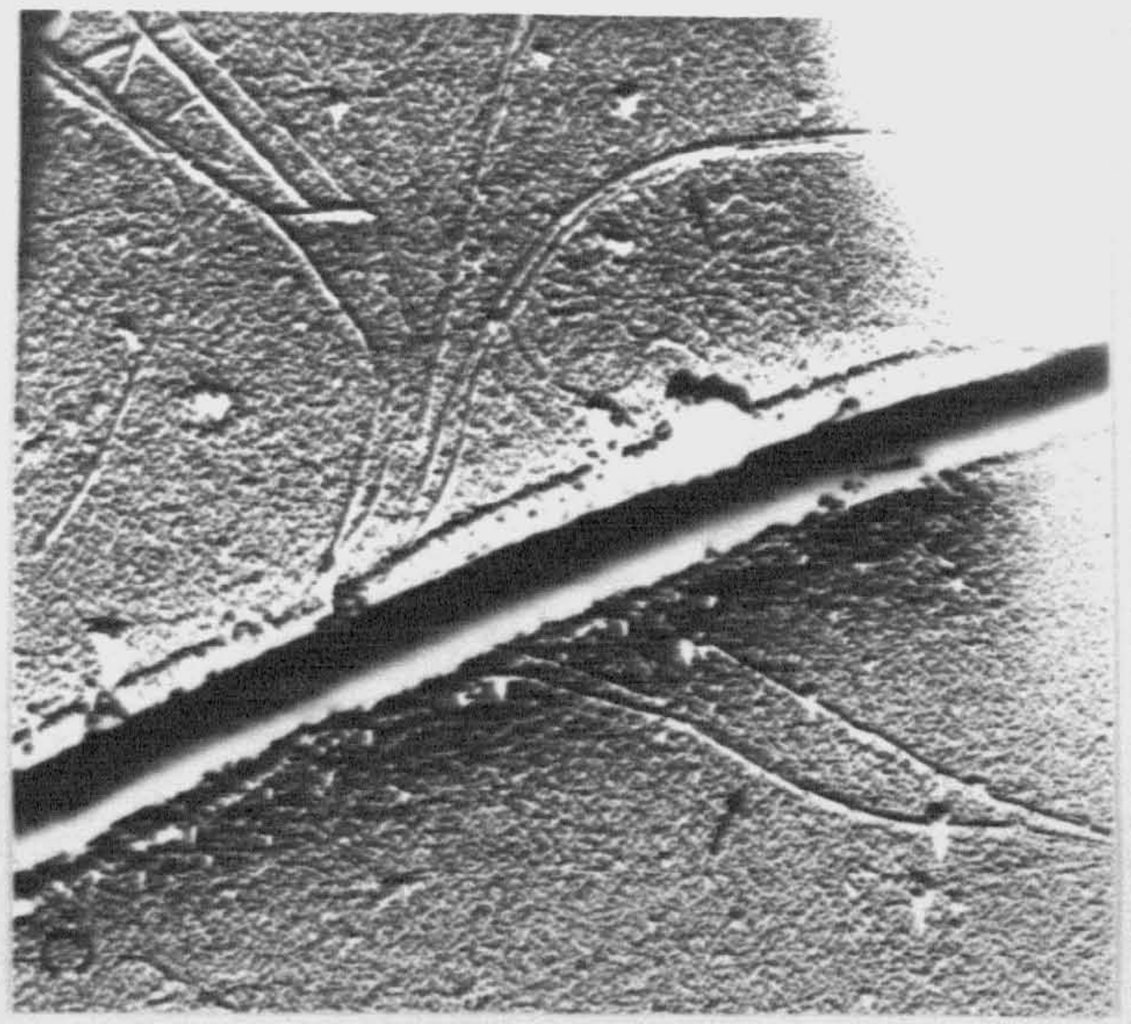
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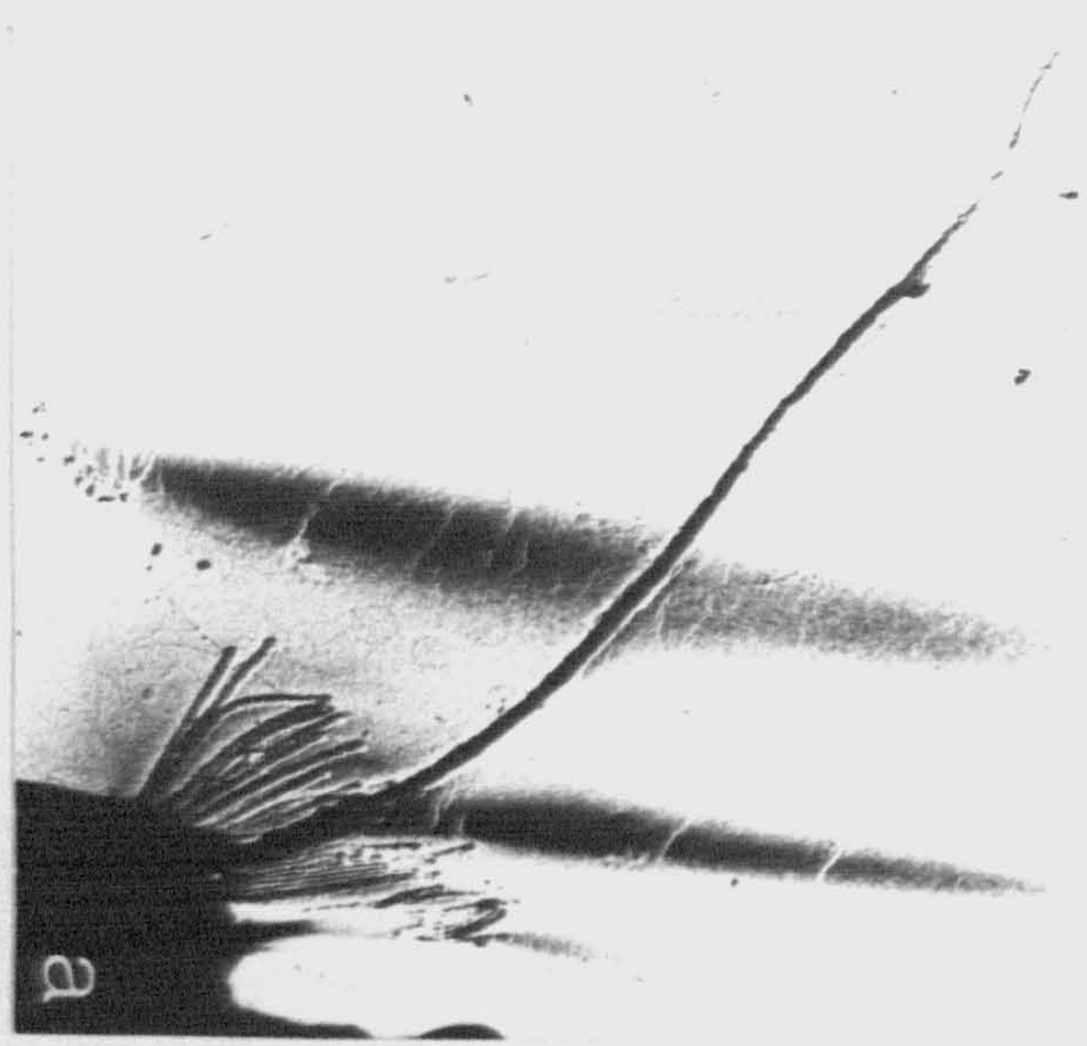
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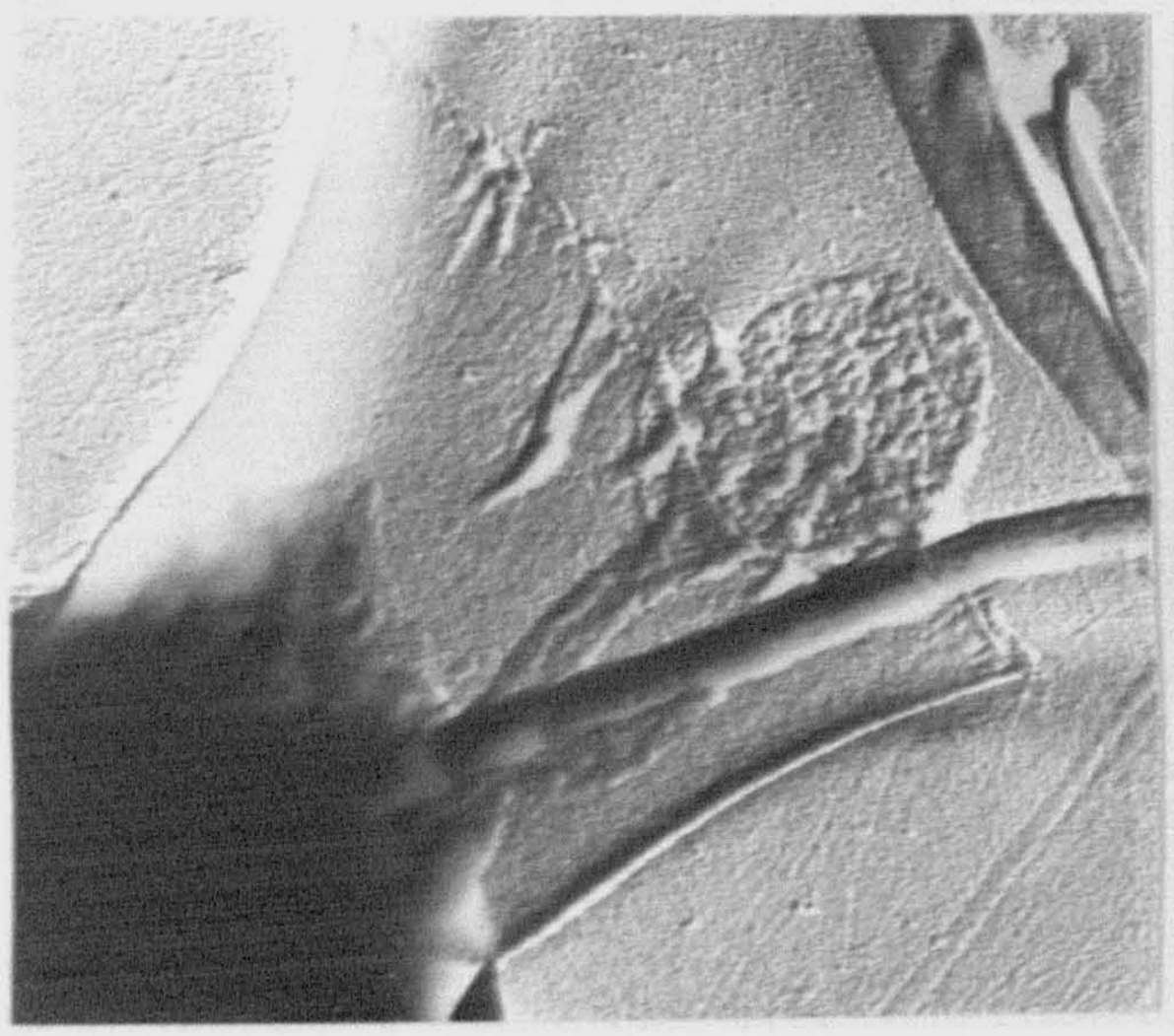
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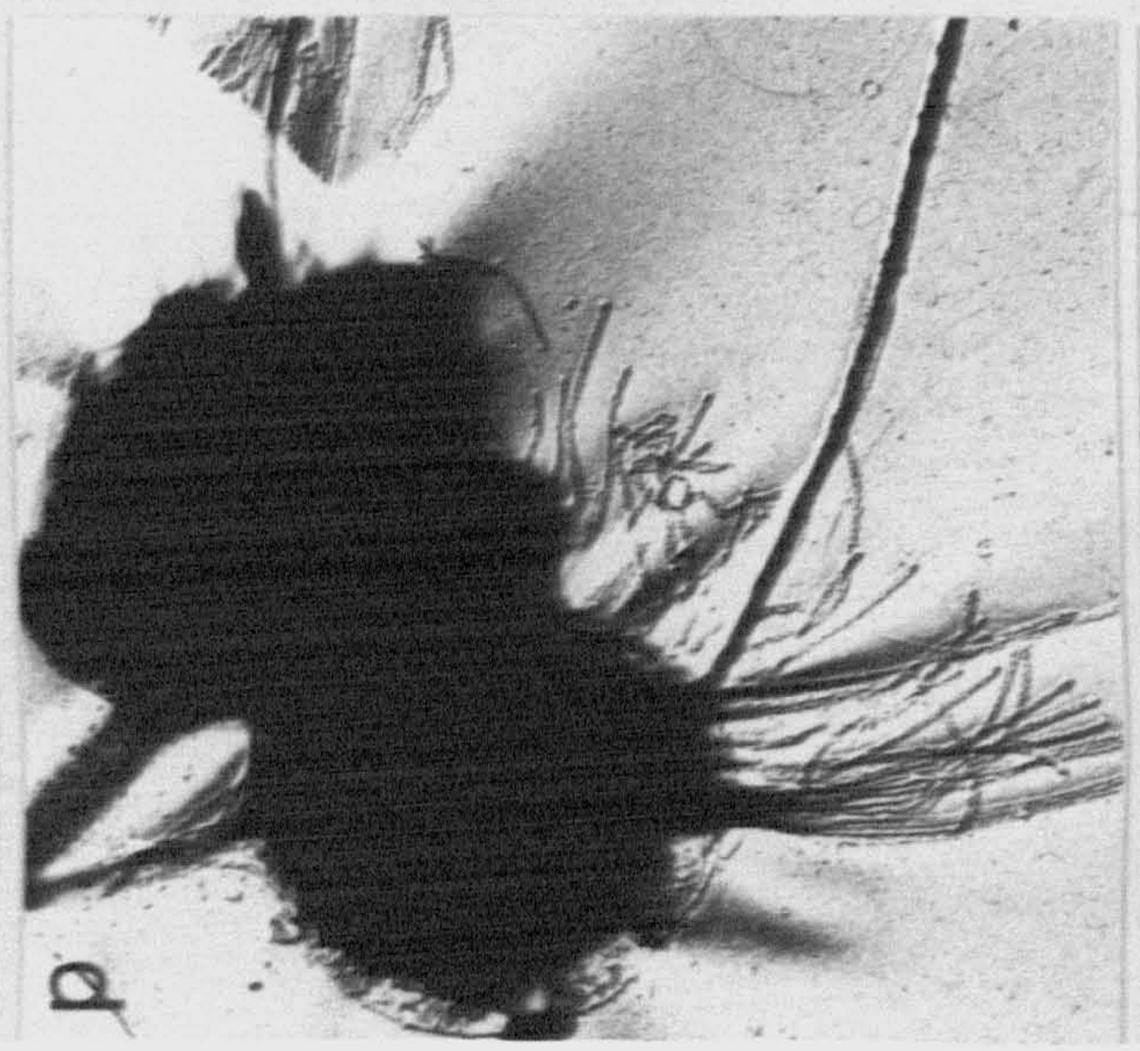
b

Salpingoeca James-Clark

Salpingoeca was established as a genus by JAMES-CLARK (1867) and SAVILLE-KENT (1881-82) discovered a large number of different forms.

The species of this genus are characterized by a vase-like chitinous lorica to which stalked or stalkless cells are attached. The cell body is mostly freely movable within the lorica and not attached permanently tothe lorica. Contractile vesicles are conspicuous, two or more in number. It inhabits salt and freshwater. Multiplication is usually by transverse,rarely by longitudinal fission and by sub-division into spores.

The Salpingoeca species occurring in Shearwater are illustrated in Fig.61. It is apparent from the same figure that the species were observed at different stages, thus making the identification quite difficult. However, it is possible that the longer species (Fig. 61a-e) is possibly Salpingoeca fusiformis Kent and the smaller one is S. frequentissima (Fig. 61h,r,t). Nevertheless to avoid misidentifications, the species occurring in Shearwater will be referred to simply as Salpingoeca spp.

All Salpingoeca spp. in Shearwater have vase-shaped lorica. The length of the lorica varies between 11 - 19μm and the width between 6 - 7μm; it appears to be amorphous (Fig. 62e,f). Small species, probably S. frequentissima, were characterized by an indentation towards the posterior end of the lorica (Fig. 61h,r). The posterior end of the lorica was

either tapered or converted into a short stalk (Fig.61a-d, p,g), by which the organisms attach themselves to algae (Fig.62d). The anterior flagellum projects from the slightly tapered anterior part of the cell body measuring 19 - 22µm and is surrounded by a collar. No evidence of hairs on the flagellum was found like that of C. botrytis (Fig. 62c), and the collar was amorphous also which is the opposite to that of C. botrytis which is made up of tentacles.

The cells body contains a conspicuous nucleus which is placed anteriorly or centrally. A number of contracile vesicles varying between 1 - 3 are usually placed posteriorly and the body also contains numerous small granules. Empty loricas were also found on some occasions (Fig.61m,n,r.s).

ECOLOGY

These colourless flagellates were conspicuous on blue-green algae and diatoms but rare on Chlorophyceae which occurred at the same time.

The occurrence of Bicosoeca lacustris, Codosiga botrytis, Salpingoeca spp. (flagellates) and Stylosphaeridium stipitatum (Tetrasporales, Chlorophyceae) on or around the colonies of Coelosphaerium naegelianum, Gomphospheria naegeliana and Microcystis aeruginosa are illustrated in Figs 63 and 64.. It is apparent that colourless flagellates are either attached to the blue-green algal cells or arrange themselves around the colonies, e.g. Fig. 63b. At the same time, Aphanizomenon flos-aquae was quite high in numbers yet was not colonised. Additionally, bacteria accumulated around the colonies of C. naegelianum, G. naegeliana and particularly M. aeruginosa; feeding on bacteria by colourless flagellates has recently been shown and hence there might be a symbiotic effect. Absence of these flagellates from Aphanizomenon flos-aquae may indicate that these flagellates cannot attach or that the necessary bacteria are not present.

The degree of the occurrence of these flagellates and S. stipitatum on the colonies of C. naegelianum, G. naegeliana and M. aeruginosa is shown in fig.65, from which it can be seen that they appeared simultaneously but remained for differing periods of time on each alga.

The degree of infection by B. lacustris on C. naegelianum in 1979 was quite high (maximum of 68%) but the occurrence was

Fig.63. The attachment of Bicosoeca lacustris, Codosiga botrytis, Salpingoeca spp. (flagellates) and Stylosphaeridium stipitatum (Tetrasporales, Chlorophyceae) to planktonic blue-green algae.

c,h,j,n B. lacustris

a,d,g,i,k,l C. botrytis

e,f,o,p,g Salpingoeca spp.

r,s,t,u Stylosphaeridium stipitatum

b showing the abundancy of colourless flagellates around the blue-green algal colony. X160

e,f,u at X860

the remainder at X450

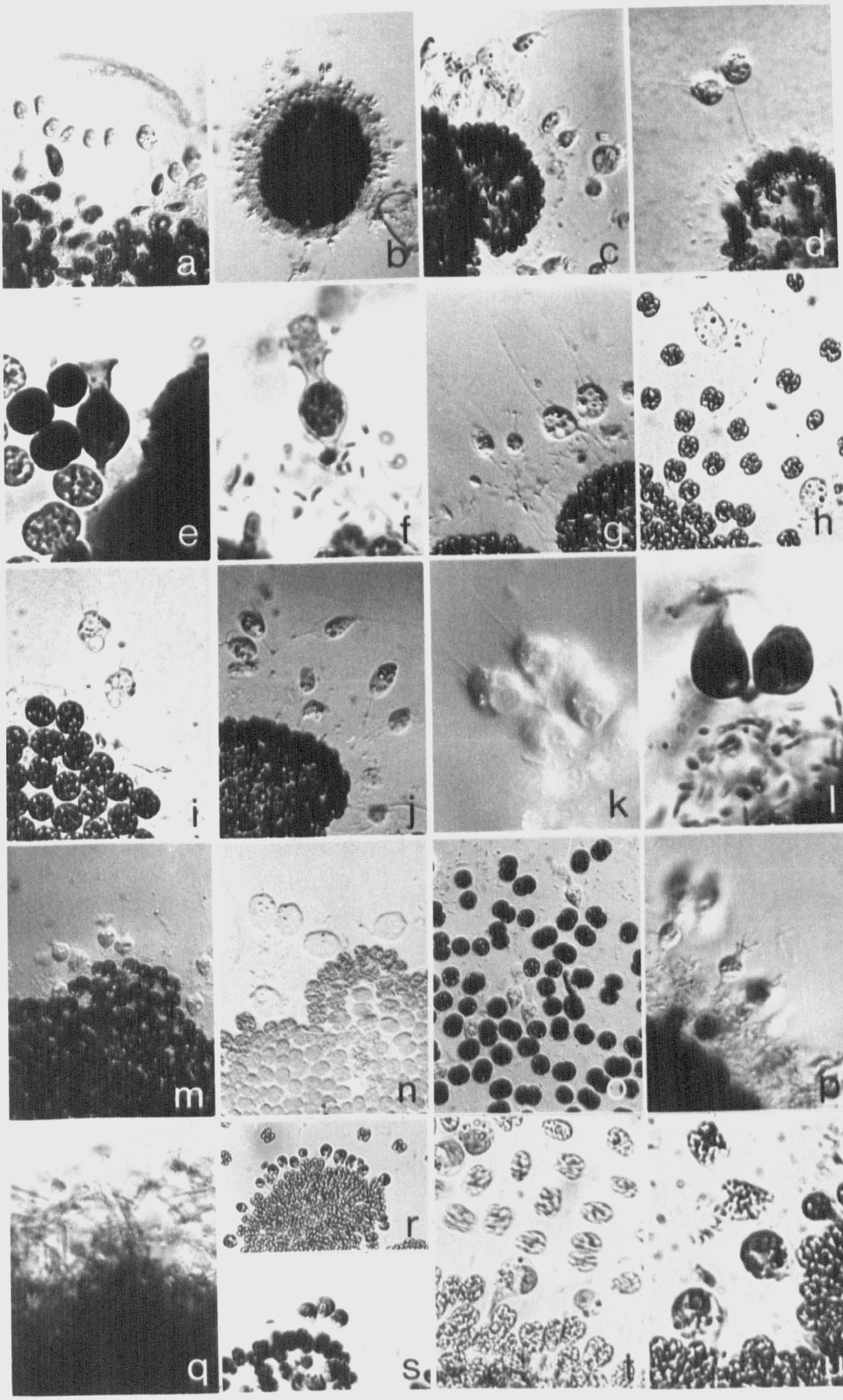
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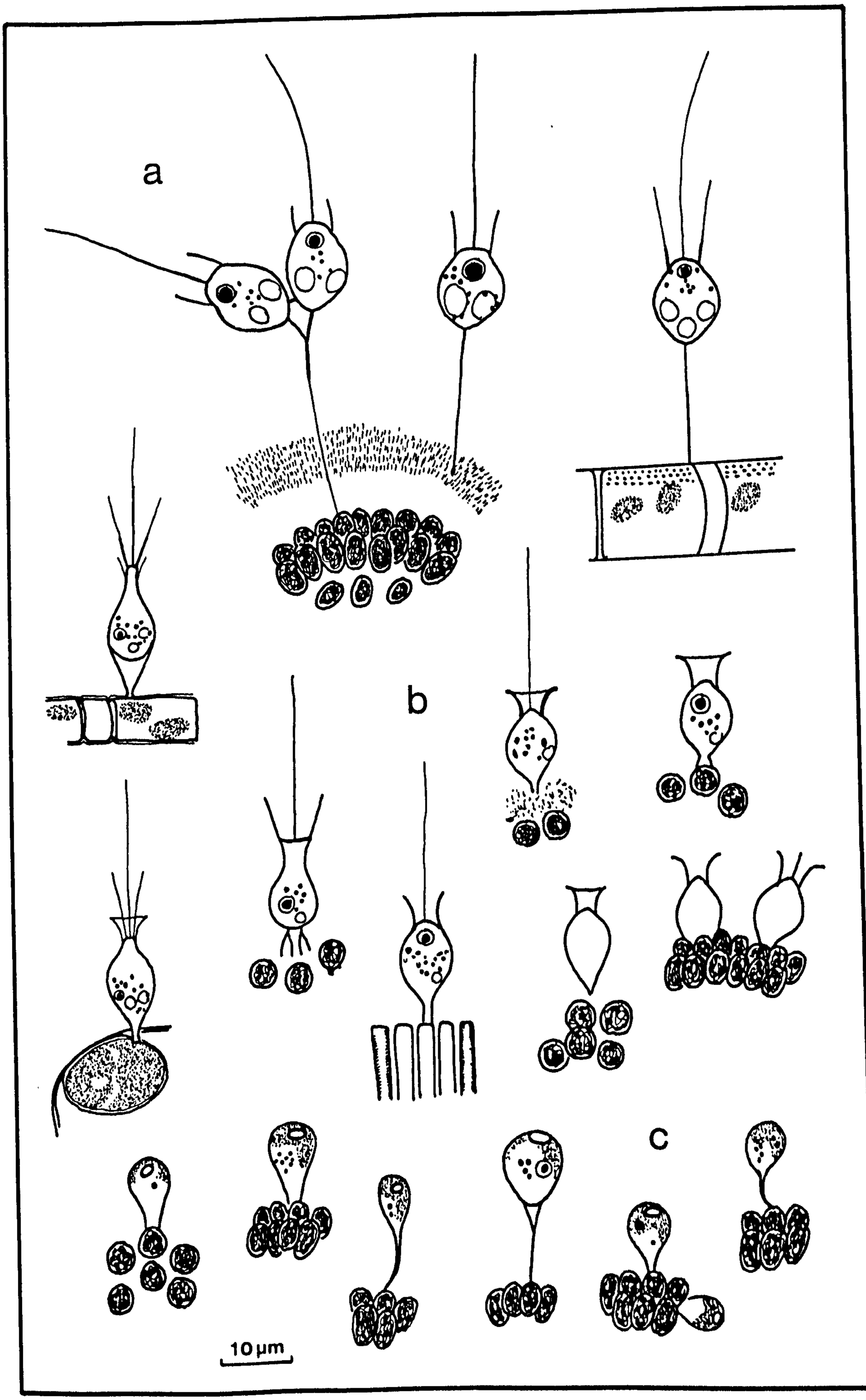
Fig.64. Occurrence of Codosiga botrytis (a), Salpingoeca spp. (b) and Stylosphaeridium stipitatum (c) on blue-green algae and diatoms.


a
b
c
10 µm

for a short time (Fig. 65a). Maximum of 21 B. lacustris were recorded on a colony but the average was 4. Attachment of C. botrytis to C. naegelianum was unimportant compared with that of B. lacustris. Maximum attachment to the colonies was 15% and average number was again 4. Salpingoeca spp. occurred in low numbers on the colonies. Maximum attachment was only 12% and the flagellate disappeared a month earlier than the other epiphytes. However, S. stipitatum showed a high degree of occurrence on the colonies and reached a maximum attachment rate of 70% before the maximum of B. lacustris was recorded on the colonies. The average number of S. stipitatum per colony was 4. All these epiphytes disappeared from the colonies of C. naegelianum simultaneously at the end of October 1979. The alga was very scarce in 1980 and no epiphytes were found.

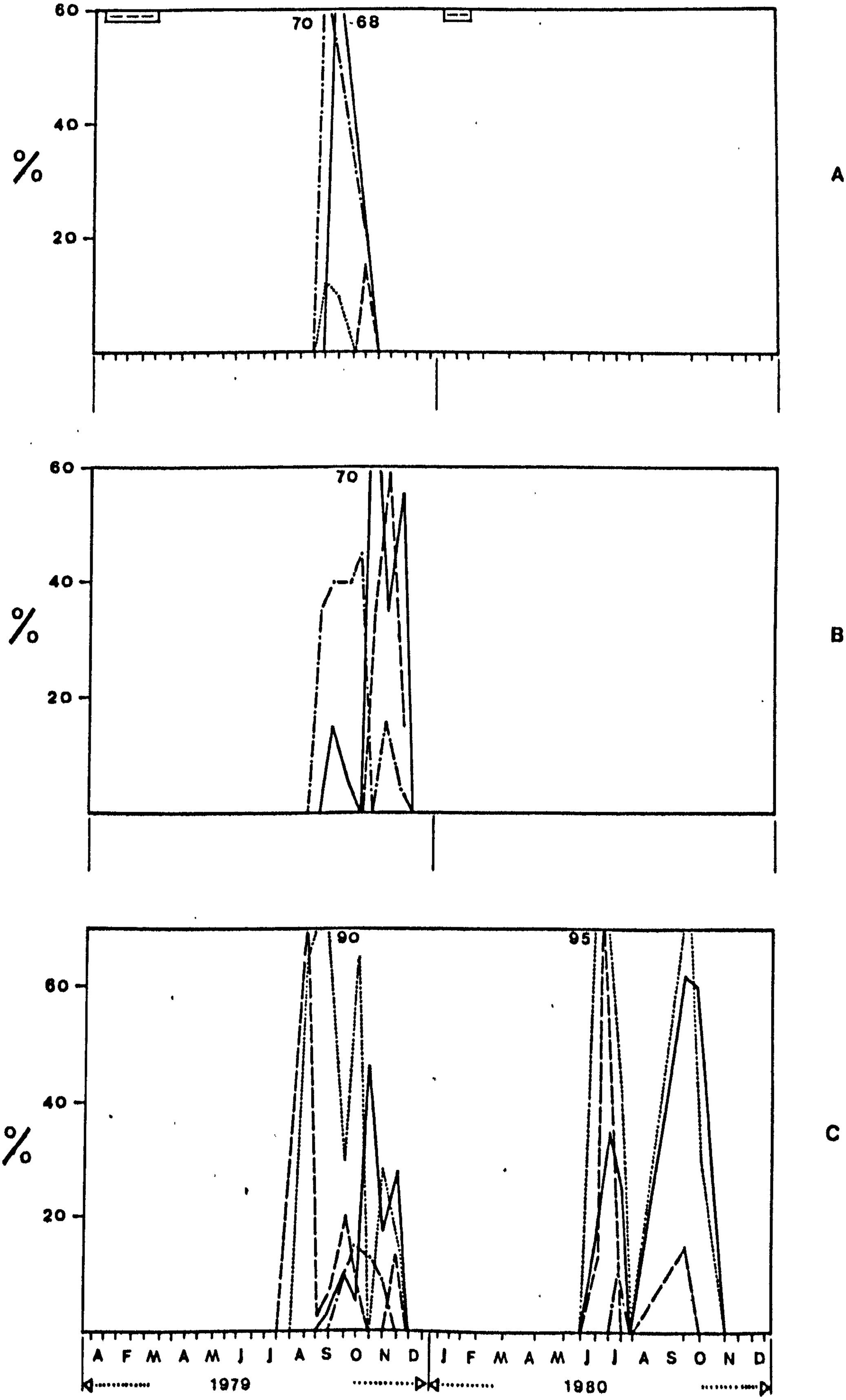
B. lacustris was also quite abundant around the colonies of G. naegeliana and reached its maximum (70%) degree of occurrence later than that on C.naegelianum. (Fig.65b). The duration of the flagellate was also longer. The average number of 4 B. lacustris was recorded per colony. The decline of B. lacustris was also sharp. Codosiga botrytis was quite abundant (average of 2 cells) on the colonies of Gomphospheria. Presence of Codosiga on the colonies was detected much later than that of Bicosoeca and the maximum number of the former was later than the latter. It was quite interesting that attachment of Salpingoeca to Gomphospheria was really scarce. S. stipitatum occurred densely on Gomphosphaeria as on Coelosphaerium. The number of Stylosphaeridium increased rapidly and reached a

Fig.65. Seasonal distribution of B. lacustris, (_____), C. botrytis (_ _ _ _ _), Salpingoeca spp. (.....), and S. stipitatum (_._._._).

A, on Coelosphaerium naegelianum

B, on Gomphosphaenia naegeliana

C, on Microcystis aeruginosa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90
95
A F M A M J J A S O N D J F M A M J J A S O N D
1979
1980
C

maximum of 60% within a very short time. However, disappearance of the species was equally quick and was synchronous with the disappearance of the colourless flagellates. The average number of 4 Stylosphaeridium per Gomphosphaeria colony was recorded; it was absent in 1980.

Occurrence of Bicosoeca lacustris around the colonies of Microcystis aeruginosa in 1979 was not as conspicuous as around Coelosphaerium and Gomphosphaeria (Fig.65c). It appeared much later than Codosiga or Salpingoeca and reached a maximum (46%) whilst the latter flagellates were declining in numbers. An average number of 4 Bicosoeca per colony occurred. However, Bicosoeca achieved higher numbers in 1980 and the maximum attachment rate was 62%. In addition, the duration of presence of the flagellate was also longer in 1980.

Codosiga botrytis was quite abundant on Microcystis in 1979 and 1980 and showed a more or less similar pattern of distribution in both years. The occurrence of the flagellate on Microcystis was earlier than observed on Coelosphaerium and Gomphosphaeria. Maximum attachment rate was 70% in both years which was followed by a sharp decline in the numbers of the flagellate. Average number of Codosiga around the colonies was 7.

The occurrence of Salpingoeca spp. on Microcystis was the most conspicuous of all these flagellates. Salpingoeca app. were quite abundant in both years (Fig.65c); 90 - 95% occurrence, indicating that Salpingoeca occurred on almost every single colony of Microcystis. These flagellates were present on the alga for a long period with high numbers and showed two characteristic maxima in each year. Average number of 10 Salpingoeca per colony

Fig. 66. Occurrence of Bicosoeca lacustris, Codosiga botrytis and Salpingoeca spp. on planktonic diatoms.

a,b,f B. lacustris on Melosira ambigua

e on M. granulata

h,i,j on Asterionella formosa

k,l on centric diatoms

n,o,r,s,t on Fragilaria crotonensis

d C. botrytis on M. ambigua

g on M. granulata

m on F. crotonensis

c Salpingoeca sp. on M. ambigua

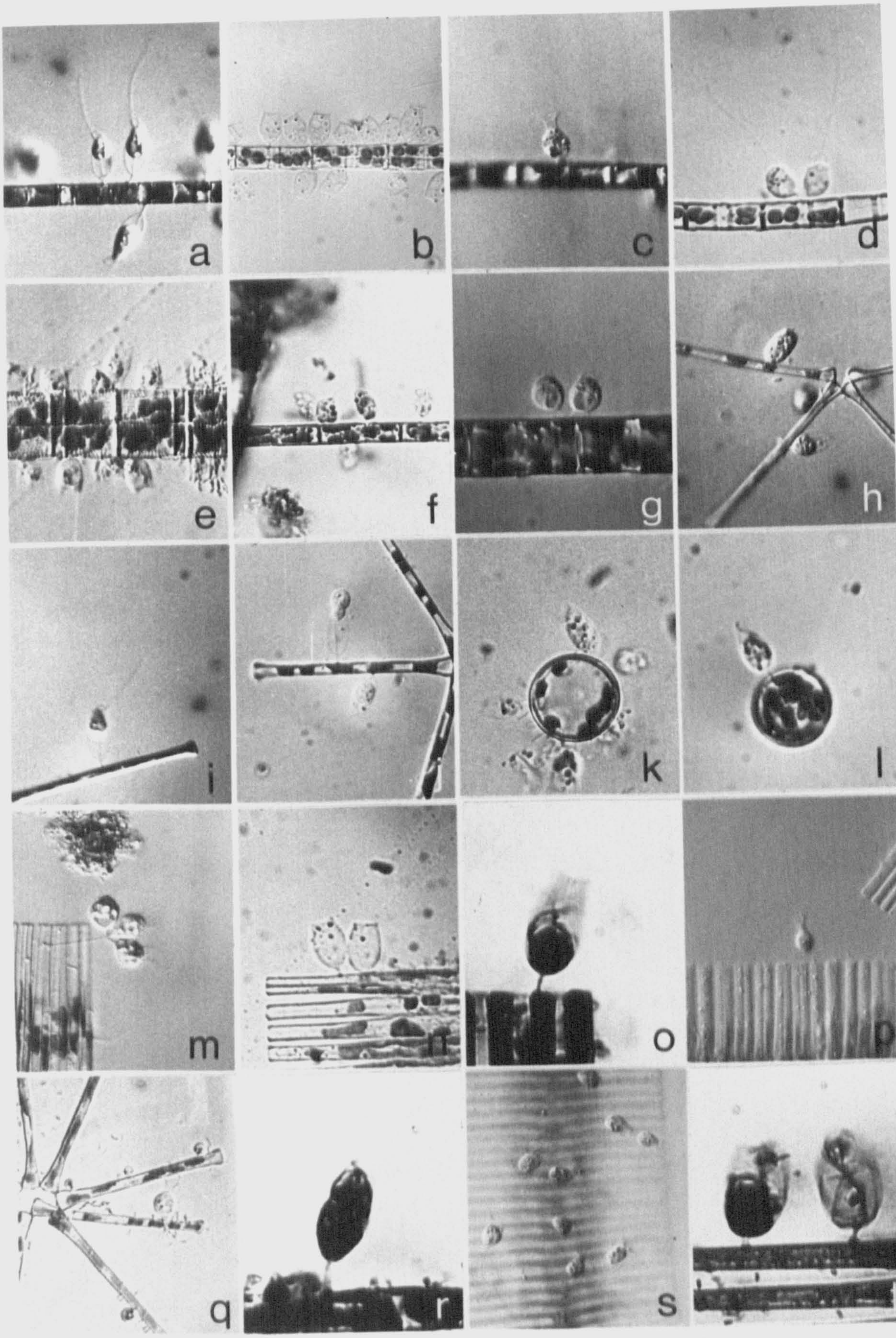
p on F. crotonensis

g on A. formosa

o,r,t, at X860

remainder at X410

Note the crowded population of Bicosoeca on M. ambigua (b), M. granulata (e) and on F. crotonensis (s).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was recorded.

However, the occurrence of Stylosphaeridium on Microcystis was surprisingly low compared with that on Coelosphaerium and Gomphosphaeria. This suggests that Stylosphaeridium shows a degree of selectivity. On average only one cell occurred per Microcystis colony.

Bicosoeca, Codosiga and Salpingoeca were also quite conspicuous on the planktonic diatoms. In general Bicosoeca lacustris was the dominant flagellate. The occurrence of the flagellates on planktonic diatoms is illustrated in Fig.59 and 66. It is apparent from these figures that the flagellates settle down on the cells of the diatoms and can live on the cells even after the cells die.

The occurrence of Bicosoeca lacustris on Fragilaria crotonensis was the most conspicuous of the populations occurring on diatoms. Bicosoeca usually appeared on Fragilaria during the summer - autumn period coinciding with either increasing or high numbers of the diatom. 18 Bicosoeca was determined as an average number on per filament of Fragilaria, indicating the high degree of occurrence.

Codosiga botrytis and Salpingoeca spp. only occurred on Fragilaria during the summer - autumn period. Their occurrence was less important than that of Bicosoeca. Average number of 3 individuals on per filament was recorded.

Occurrence of Bicosoeca on Asterionella formosa was unimportant since the highest maximum attachment was only 14% (Fig.67b). Seasonal distribution of Bicosoeca on Asterionella showed that the flagellate could be found on Asterionella in

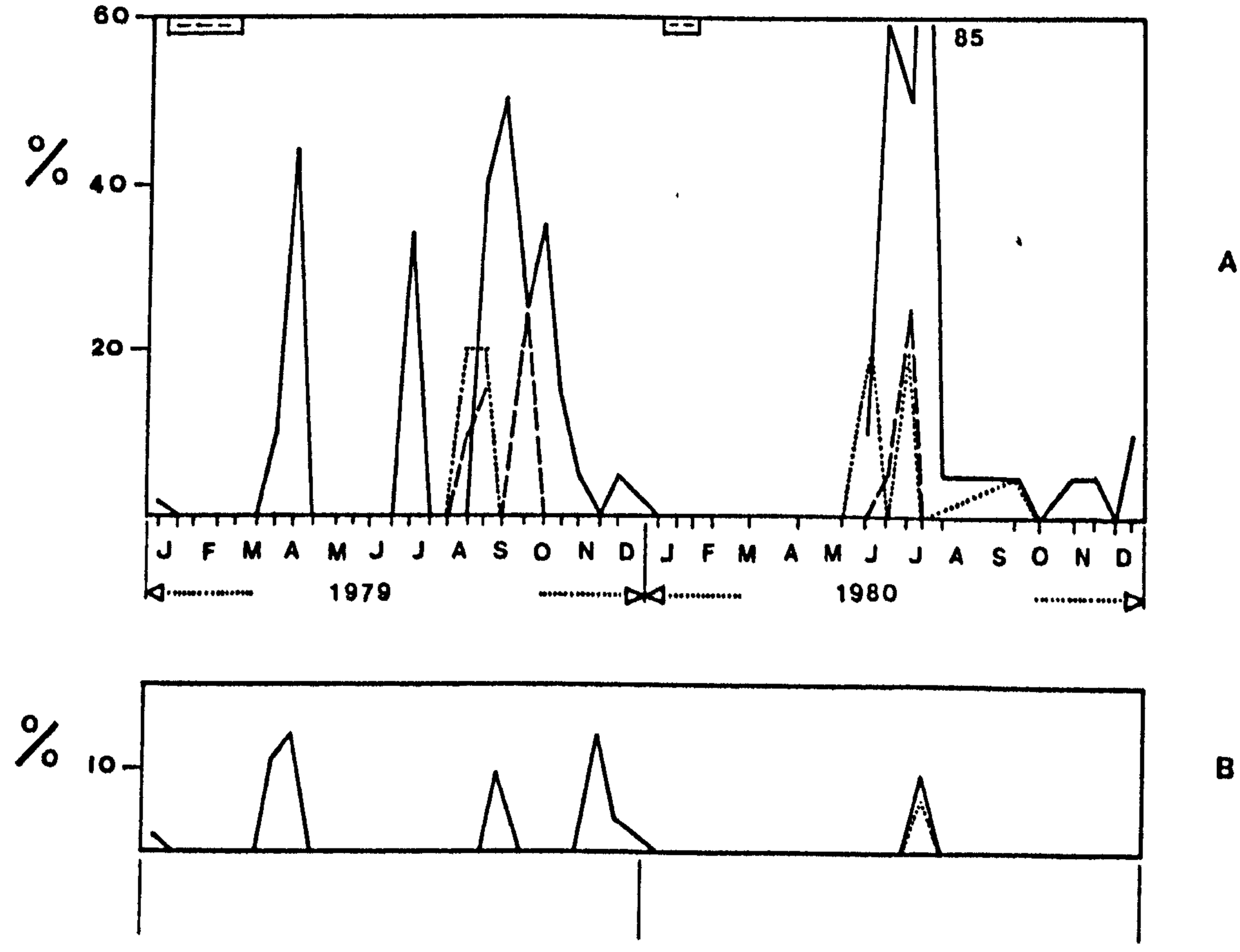
Fig. 67. Seasonal occurrence of Bicosoeca lacustris (_____), Codosiga botrytis (_ _ _ _) and Salpingoeca spp. (.....).

A, on Fragilaria crotonensis

B, on Asterionella formosa

C, on Melosira ambigua

D, on Melosira granulata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every season but with small numbers. Average number of 2 Bicosoeca were recorded per Asterionella cell.

Bicosoeca was found also on the filaments of Melosira ambigua and M. granulata in the same periods (Fig.67c,d). Attachment of the flagellate to the diatoms was usually for a short time and observed in every season except winter. However, Bicosoeca was irregularly present on M. granulata for a long period during autumn 1979. Maximum attachment of 80% and 85% was recorded in the case of M. ambigua and M. granulata respectively. Average number of 4 and 13 Bicosoeca per filament was recorded for M. ambigua and M. granulata respectively. Attachment of Codosiga and Salpingoeca to the filaments of Melosira spp. was found during autumn as well as summer. However, only Codosiga occurred on M. granulata while Salpingoeca was absent. Maximum attachment of 35% for Codosiga was recorded on M. granulata. Salpingoeca was only found on M. ambigua and this coincided with its occurrence on blue-green algae.

The flagellates were also occasionally found free or attached to green algae. (Fig.68). The occurrence of the flagellates on the remains of planktonic algae was also observed (Fig.68b,e) and occasional high numbers of Bicosoeca lacustris were recorded on Oocystis and Staurastrum (Fig.68k,p).

In conclusion, the present data would suggest that the colourless flagellates, Bicosoeca, Codosiga and Salpingoeca and the green epiphytic algae Stylosphaeridium showed a characteristic seasonal periodicity and some degree of host specifity. Their regular occurrence in summer - autumn clearly indicates a restriction by certain ecological factors; silica has been

Fig. 68. (a - d) Codosiga botrytis, free (a) or attached to debris (b,d).

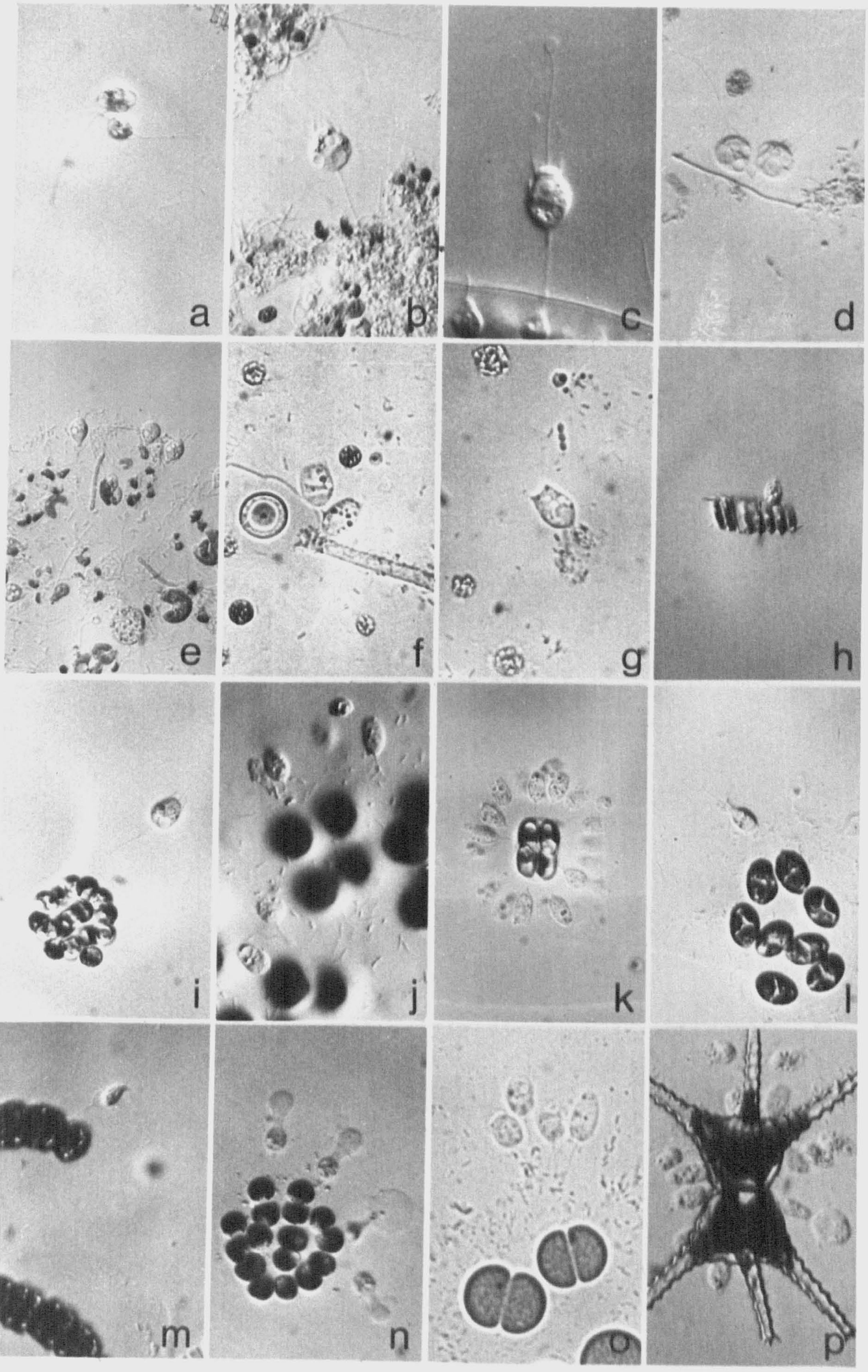
(e) Salpingoeca sp. on remains of Kirchneriella obesa.

(f, g) Bicosoeca lacustris on debris.

(h - p) attachment of these flagellates to planktonic blue-green and green algae.

Nore: abundancy of B. lacustris on Oocystis lacustris(k) and on Staurastrum (p).

All pictures at X450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implicated in the formation of the theca but further studies are required before any relationship is revealed.

Summary and conclusions

Bicosoeca lacustris, Codosiga botrytis and Salpingoeca spp. displayed a distinct seasonal distribution on or around blue-green algae and diatoms whilst the epiphytic green alga Stylosphaeridium occurred characteristically only on blue-green algae.

Numbers of these epiphytes differed from alga to alga indicating a degree of host specifity.

Their occurrence in high numbers is restricted to summer - autumn periods along with high populations of algal hosts.

The effect of the occurrence of epiphytes on the algal hosts require experimental studies. Symbiotic effects almost certainly occur.

CHAPTER 5.

DISCUSSION

The aims of the present work were to study the epiphytic organisms associated with the phytoplankton of Shearwater and assess their effect on the population dynamics. In order to commence a study of the epiphytes it was necessary to simultaneously establish the annual cycle of the phytoplankton. In addition if the deleterious or stimulatory effects of the epiphytes are to be studied then the influence of all the physico-chemical variables needs to be considered. Thus a three-pronged approach had to be used.

Taking only the commonest algae in the phytoplankton of Shearwater there are some 32 species to be considered, of which 15 are Diatoms, 12 are Chlorophyta, 5 are Cyanophyta. Their seasonal cycles turned out to be complex. Unlike many oligotrophic bodies of water where the periodicity of the algae is fairly simple with rapid pulses of species succeeding one another and with species confined to short periods of the year, Shearwater proved to have many species fluctuating irregularly throughout the three years investigation. The result was a very confused picture even for algae such as Asterionella and other diatoms which usually show precise growth patterns related to silica supply and its depletion. The situation may have been complicated by using a shore-based sampling site rather than the more normal central buoy position. However, if this is so, it has also given the interesting result that there is an innoculum of many species persisting in the shallow water adjacent to the shore throughout much of the year. Alternatively and equally

likely the continuous supply of nutrients in this small lake in a rich catchment area may be the cause of the almost continuous growth of so many species.

This phenomena has in fact been recorded in large lakes when the nutrient status is increased by eutrophication, e.g. the spreading of growth peaks of algae throughout the year in Lake Erie (DAVIS,1964) in recent years compared to the earlier decades of this century. It is unfortunate that the physico-chemical data often yields few clues to the algal cycles and often each year's cycle differs from the previous. Some basic patterns have however been established, but more sampling at closer intervals will be needed before correlations can be established. Under these conditions of relatively high nutrient supply it is possible that stimulatory or inhibitory effects of species upon species are playing a more important role than normal in a lake. Certainly at times of the year Shearwater has a very mixed "soup" of algae in the water column - colouring the water and forming water blooms. It is under such conditions that extracellular products might have considerable effects.

The epiphytes were studied in some detail but these also proved to be very much more complex than is normal in more oligotrophic situations. The total number is far greater than recorded here, since the bacteria were left out of this survey. Nevertheless there are 10 protozoa, 12 chytrids and 1 algae epiphytic on the phytoplankton of Shearwater. The method of study and analysis of the parasitic epiphytes (mainly chytrids) was inspired by CANTER's studies over 35 years in the English Lake District. Many common chytrids occur in

both regions suggesting that so long as the host is present the nutrient status of the water does not prevent the occurrence of the chytrid. Discovery of all the stages in the life cycle of a chytrid by observation of the natural populations is hazardous. CANTER in several publications has admitted that she has only been able to ascertain partial life cycles of some chytrids. In the present work, it was not possible to visit Shearwater (some 40 miles from Bristol) more than once every two weeks and even this was not always possible. In future studies more frequent sampling will be required to complete some observations. Nevertheless, a large amount of data has been assembled on this lowland lake to compare with the upland lakes studied by CANTER. Hardly any other lowland waters have been studied in this way in Britain and hence there is little with which to compare this study. However, it has been known from WESENBERG-LUND's early view (1905) that most phytoplankton species are "eaten" by phycomycetous fungi and this is true for Shearwater. It is surprising how little work has been undertaken on this topic in the years since WESENBERG-LUND's pioneering work. The present work has shown that the fungal attacks can drastically reduce algal populations though recovery is often equally dramatic-perhaps due to the richness of the flora and the often adequate nutrient supply so that rarely is there the double stress of parasitism and nutrient limitation. Attempts as in many of CANTER's papers and in North American work to relate the occurrence of the fungi to the ambient physico-chemical conditions have sometimes yielded similar correlations but again all too often succeeding years have complicated the picture. This aspect

and also the completion of many life cycle studies will only be satisfactorily achieved by dual culture of the algae and fungus - a goal only recently achieved by CANTER & JAWORSKI for a very limited number of co-existing pairs of organisms.

Of great interest in the present study has been the discovery of such large populations of Choanoflagellates on a wide range of phytoplankton species. These fascinating protozoa (or colourless algae?) have never been investigated from an ecological standpoint. Their effect on the algae seems to be simply to overload the cells and colonies with excess weight. However, it will be necessary to pursue these interactions in greater detail and ideally under experimental conditons. So far there is no literature on this topic. In addition, there is much still to be discovered concerning the ultrastructure of these delicate flagellates and much to clarify in their taxonomy. The present study has simply shown how widespread they are and it is inconceivable that such large populations of epiphytes have no effect on the cycling of the individual algae.

In conclusion, Shearwater has proved to be an exceptionally rich source of phytoplankton and epiphytes. It has been frustrating in that the algae and epiphytes have rarely shown precise patterns and succeeding years whilst showing certain basic similarities have often provided contrary data. Nevertheless, in such an ecological survey, it is the reality and there must be underlying causes even if these have proved difficult to discern.

This study has been involved with the occurrence and seasonal distribution of the epiphytic flora of the phytoplankton

and this is really only the initial survey part of their ecology. The chemical cycles involved due to the breakdown of first the algae and then presumably the fungi/protozoa themselves and the resulting nutrient cycling is still very much a closed book in limnology. This aspect, however,is a vital one to the understanding of phytoplankton ecology since in waters such as Shearwater the rapid internal cycling of nutrients such as silica may largely explain the almost continuous production of diatoms, e.g. BAILEY-WATTS (1976a) has shown that over a one-month cycle in Loch Leven the release of silica from the sediments and from the dissolution of diatom cells was more important than the external supply. As in this study of Shearwater another paper by BAILEY-WATTS (1976b) showed that diatoms increased at times when silica was increasing but at others when it was decreasing. It is interesting that Loch Leven also is a shallow eutrophic body of water. In addition, some of the algae of the phytoplankton of Shearwater (e.g. Pediastrum) have been shown to have heterotrophic tendencies (BERMAN et al. 1977) and this could account for their extended growth over periods when inorganic nutrients might be limiting. Such a situation has never been tested experimentally in a limnological study, only in laboratory cultures.

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